PrepLadder

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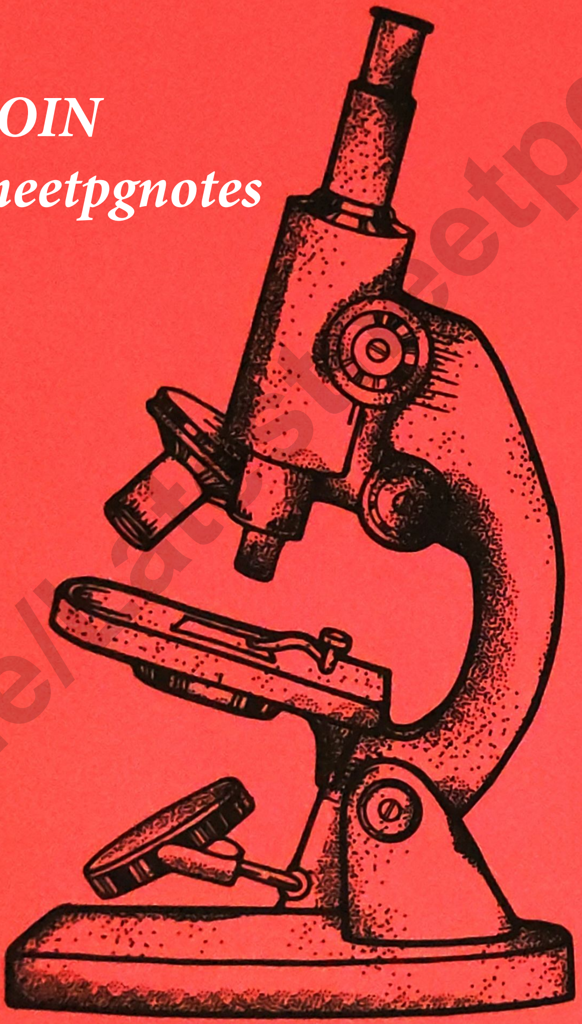

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MICROBIOLOGY

Based on video lectures by
Dr. Sonu Panwar

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Unit: 1 INTRODUCTION

Chapter 1

Introduction to Microbiology

GRAM POSITIVE COCCI

All Cocci are Gram +ve Except

- Meningococci
- Gonococci
- Veilonella
- Moraxella

GRAM NEGATIVE BACILLI

All Bacilli are gram -ve Except

M → Mycobacterial species
A → Anthracis bacillus
C → Clostridium species
D → Diphtheriae corynebacterium (Pathogonomic)
O
N → Nocardia
A → Actinomyces
L → Listeria
D → Diphtheroids (Normal commensal of throat)

STAINING

00:12:40

- Gram staining } differential
- Acid fast staining [ZN staining] } staining
- Bipolar staining

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1. GRAM STAINING [DISCOVERED BY CHRISTIAN GRAM]

Sequence of Bacterial Smear

Methyl → Methyl violet /crystal violet/gentian violet → primary stain

↓

G → Gram's Iodine → Mordant (fixer)

↓

A → Acetone [for 2-5 sec] → Decolourizer

↓

S → Safranin → Counter stain

Differentiates b/w gram positive & gram negative micro organism

- Gram positive → Purple/violet
- Gram negative → Pink

FACTORS GOVERNING GRAM STAINING

a. Cell wall thickness [Peptidoglycan Layer Thickness]

- Most important factor governing gram staining
- Thick peptidoglycan layer → Gram positive feature
- Thin peptidoglycan layer → Gram negative feature

b. Dye - iodide complex

- Strong → Gram positive
- Weak → Gram negative

c. LPS [Lipopolysaccharide]

- Feature of gram negative microorganism
- Dissolved by acetone

Eg:

- Gram +ve (S. aureus)
- Gram +ve Cocci

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Bacterial smear

↓

M. Violet } Dye-I-complex

↓

Grams I_2

↓ Acetone (Decoloriser)

Safranin

i) Thick PG layer

ii) Strong Dye-I-Complex

→Gram +ve Cocci (S. aureus): Purple to violet

→ Gram –ve (E. coli)

Bacterial smear

↓

M. violet

↓

Grams I_2

↓ Acetone

Safranin

i) Thin PG layer

ii) Weak Dye-I-Complex

iii) LPS present

→Gram –ve : Pink

d. Protoplasm

- Strong → Gram positive [more acidic]
- Weak → Gram negative [less acidic]

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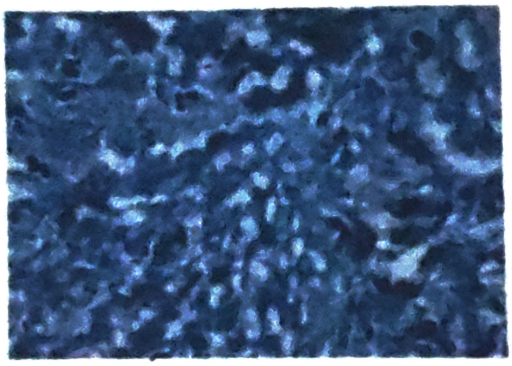
2. ACID FAST STAINING [ZIEHL NEELSEN STAINING]

00:24:30



ZN Stain

- Gram Stain
- Acid fast stain

} Differential stain

- GS differentiates between Gm +ve & Gm -ve
- AF differentiates between Acid Fast & Non Acid Fast
- Fastness → ability to resist decolorisation

↓

Due to MYCOLIC ACID [Content]

- Mycobacterium tuberculosis → 20% acid fast
- Mycobacterium Leprae → 5% acid fast
- Nocardia → 1% acid fast
- Bacterial spores
- Cryptosporidium parum
- Cyclospora
- Isospora

  These are

  ↓

  Coccidian Parasites

  ↓

  Diarrhea in HIV +ve
- Hooklets of eggs of T. Saginata
- Scolex of E. Granulosis
- Spermatic head
- Legionella micadedi

} 0.25-1% acid fast

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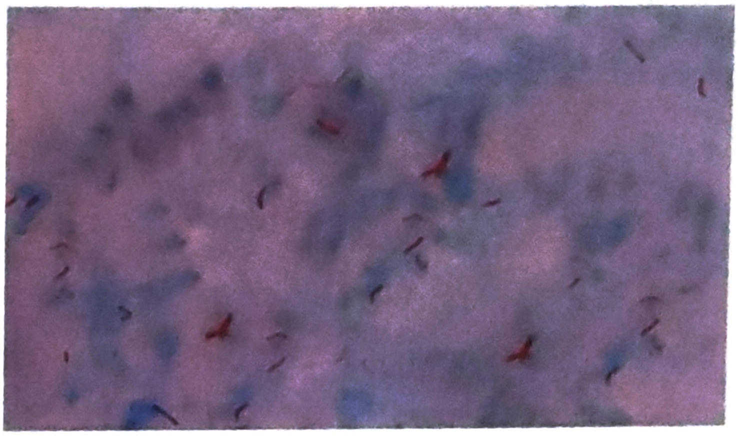
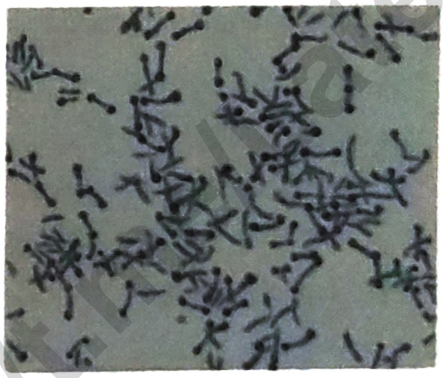
KINYOUN METHOD OF STAINING



KINYOUN STAINING

- Modification of ZN staining
- Cold method of staining (phenol concentration is ↑sed for fixation)
- Heating part is not done

3. Albert Staining 00:32:21



Chinese Letter Pattern

Albert Stain A	Albert Stain B
G- Glacial Acetic Acid M – Malachite Green T – Toluidine Blue Stains : C. Diptheriae - Green - Cuneiform arrangement/ Chinese letter pattern	I_2 KI

4. BIPOLAR STAINING [Safety Pin Appearance] 00:35:01

- Poles are stained, remaining spared

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- Motility Names
    - T → Tumbling → Listeria monocytogenes
    - D → Darting → V. Cholerae
    - S → Swarming → Proteus
- Falling leaf like → Giardia Lamblia [4 pairs of flagella]
- Slow form of swarming [stately] → clostridium
- Gliding motility → mycoplasma
- Treponema pallidium → cork screw motility
- Borrelia → lashing motility
- Spirochetes have ENDOFLAGELLA
    - Arise from periplasmic space
- Twitching motility →Trichomonas vaginalis
- Rotatory motility →B. coli (ciliate)
- Toxoplasma → Actin Cytoskeleton (Gliding)

METHODS TO DEMONSTRATE MOTILITY

1. Hanging drop method
2. Semi solid agar method [Mannitol motility agar] [mc used]
3. Dark ground microscopy
4. Phase contrast microscopy

CULTURE MEDIA 01:07:51

Culture Media			
Simple media		Complex media	
NA	NB	Enriched media	
(Nutrient Agar)	(Nutrient Broth)	Solid	
Peptone, Meat extract Agar : 2% Solidifying Agent	- Liquid	Eg: BA Blood Agar	CA Chocolate Agar
		5-10% RBC lysis ↓ X & V (Haemin) (NAD) ↓ Chocolate color (chocolate agar)	

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ENRICHMENT MEDIA

- Always liquid culture media
  - Have stimulating effect on bacteria to grow
  - Inhibits its competitors [usually commensal organism]
- Examples
  - Selenite-F-broth → for Salmonella, Shigella
  - Tetrathionate broth → for Salmonella

SELECTIVE MEDIA

- Mostly solid
  i) Wilson & Blair → S. typhi
  ii) Potassium tellurite agar → C.diphtheria
  iii) MSA → S. aureus

Indicator, Differential & Selective:

- Indicates the color change
- Differentiates between lactose fermenting & Non lactose fermenting organisms
- Selective for gram –ve bacilli
- Mac-Conkey agar (It is also indicator & selective media)

Contents:
P – Peptone
L – Lactose
A – Agar
N – Neutral red (Indicator)
T – Taurocholate sodium (selective agent)

- LF → changes colour to Pink
- NLF → No change

Organism which ferments Lactose:

- E. coli
- Klebsiella
- S. Sonnei

CLED

(Cystine Lactose Electrolyte Deficient Media)

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CLED >> Mac-Conkey agar

↓

- Non selective
- Prevents swarming of proteus
- Differential
- Indicator
- Non selective

TRANSPORT MEDIA

- Maintains viability but no growth

Venket Raman MEDIA

- V. cholerae

Cary Blair Media

- V. cholerae

Pike's media

- S. Pyogenes

Stuart's Amie's Media

- Gonococci & meningococci

Sach's Buffered Glycerol saline

- Shigella, Salmonella

ANAEROBIC MEDIA

- Robertson Cooked Meat Media (RCM)

Clostridium sps.

- Proteolytic
  ↓
  Turns meat black
  Eg. Cl. tetani
- Sacchrolytic
  ↓
  Turns meat pink
  Eg. Cl. perferingens

- Thioglycolate broth
- Sugar Media
  - Andrade's indicator
  - Used to study fermentation of bacteria

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- Peptone water + culture growth + glucose powder

↓

Colorless

↓

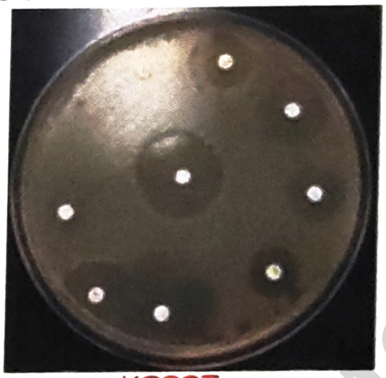
Pink

↓

Organism fermented Glucose

ANTIBIOTIC SENSITIVITY TESTING

- Done on Mueller Hinton Agar (MHA)
- Kirby Bauer Disc Diffusion Testing (KBDDT)
  - Commonly used



KBDDT

- Stoke's method of antibiotic testing
  - Better method, as controls are used

CULTURE MEDIA

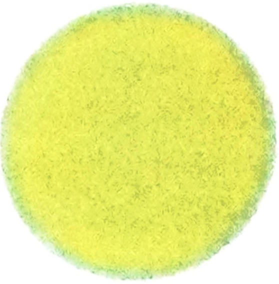
NUTRIENT AGAR

Media Type: Basal or Simple media.

Main constituents: Nutrient broth and 2% agar

Method for sterilization: Autoclaving

Color of prepared medium: Light yellow colored clear to slightly opalescent gel forms in Petri plates.



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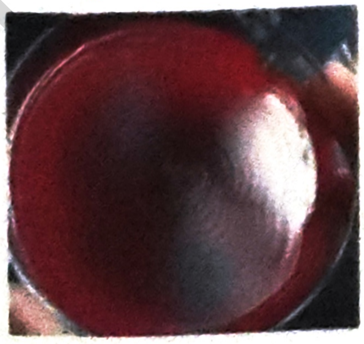
Intended use:

- It serves as the base for the other media preparation
- Isolation of non-fastidious organism
- NA slant is used for short term preservation of the bacteria.
- Performing serological and biochemical tests.
- It is used to study bacterial colony morphology and demonstrating the swarming & pigment production.
- Semisolid NA (0.2-0.4%) is used for demonstration of bacterial motility and preservation of bacteria. Additional of 6% in NA makes it firm agar that prevents swarming.

Growth of P. aeruginosa on NA

Growth of S. aureus on NA

Swarming of proteus on NA



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Motility in Semisolid NA

BLOOD AGAR

Media Type: Enriched media

Main constituents: 5-10% Sheep Blood, Nutrient Agar

Method for sterilization: Blood is added in autoclaved NA at 45 degree C.

Color of prepared medium: Cherry red colored opaque medium.

Intended use:

1. Routine culture media for isolation of fastidious organisms in Laboratories.
2. Used in demonstration of Hemolysis

- Alpha (partial) hemolysis: Some bacteria can reduce haemoglobin to methaemoglobin which produces a greenish zone around the colonies.

  Examples are Viridans streptococci and S. pnuemoniae.

- Beta (complete) hemolysis: Bacteria is surrounded by clear,colourless zone where the RBCs have been lysed and the Hb destroyed to a colorless compound.

  Examples are S. aureus, Streptococcus species etc.

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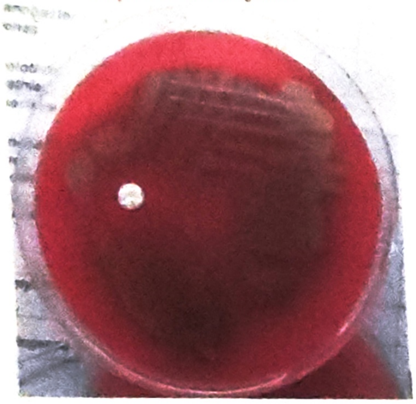
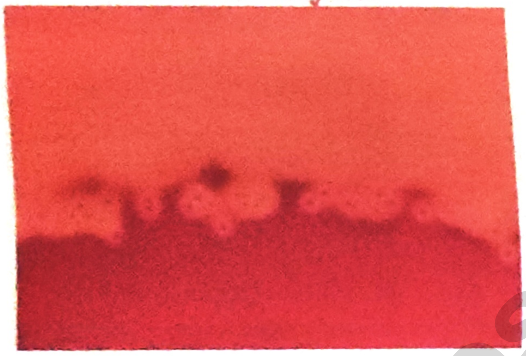
- Gamma haemolysis: There is no haemolysis

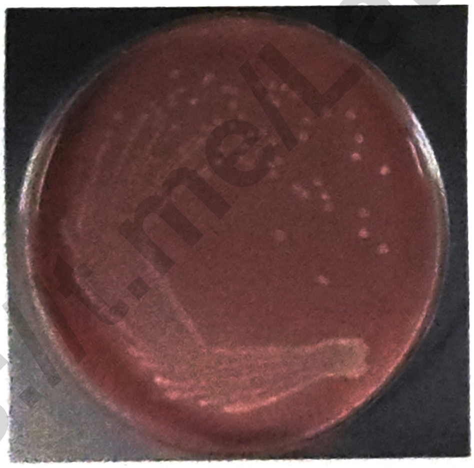
Alpha Hemolysis:



Beta Hemolysis:



CHOCOLATE AGAR



Media Type: enriched media

Other Name: Heated Blood agar

Main constituents: 5-10% Sheep Blood, Nutrient Agar

Method for sterilization: Blood is added in autoclaved NA at 70° C

Color of prepared medium: Chocolate brown

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Intended use:

- It is more nutritive than Blood agar.
- H. influenza only grows on CA as it requires X (hemin) factor and V (NAD) factors for their survival, which are only present in CA
- Isolation of Neisseria

Mac CONKEY AGAR

Media Type: Differential or Indicator media or low selective media.

Main constituents: Lactose, Peptone, Neutral red indicator, Taurocholate

Method for sterilization: Autoclaving

Color of prepared medium: Red with purplish tinge coloured clear to slightly opalescent gel forms.

Intended uses

- It is used for isolation of gram negative bacilli
- It differentiate Lactose Fermenting (LF) bacteria (E. coli, Klebsiella) from Non LF bacteria (NLF).
- LF Bacteria- Pink colored colonies
- NLF bacteria - Pale or colorless colonies

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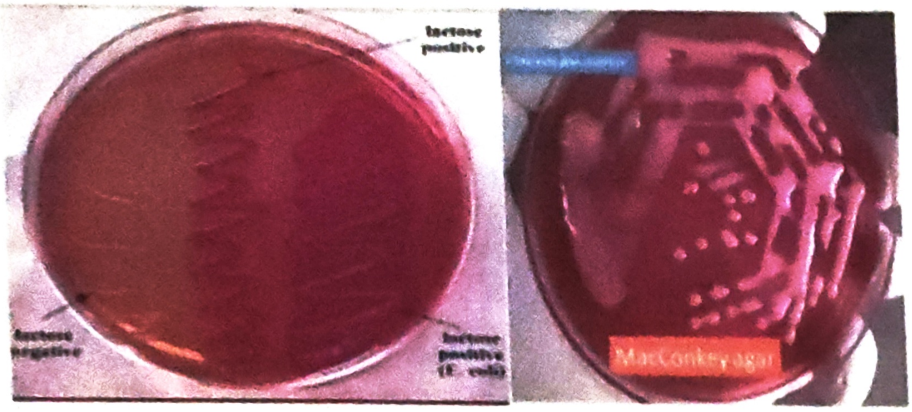
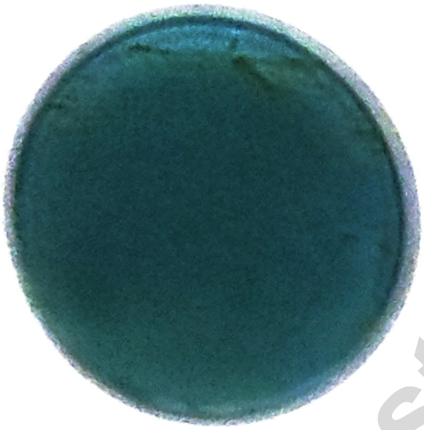


CYSTINE LACTOSE ELECTROLYTE DEFICIENT AGAR (CLED)



Media Type: Differential or Indicator media

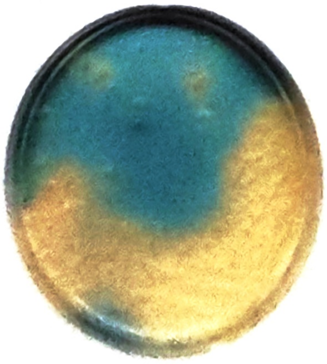
Main constituents: Cystine, Lactose, Peptone, Bromo Thymol Blue indicator

Method for sterilization: Autoclaving

Color of prepared medium: Blue-Green colored clear to slightly opalescent gel forms

Intended use:

- It is used for isolation of uropathogens
- It differentiates LF bacteria (yellow colonies) from non LF bacteria
  - It is less inhibitory to gram positive bacteria
  - Prevents swarming of Proteus



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LOWENSTEIN JENSEN MEDIUM

Media Type: Selective media

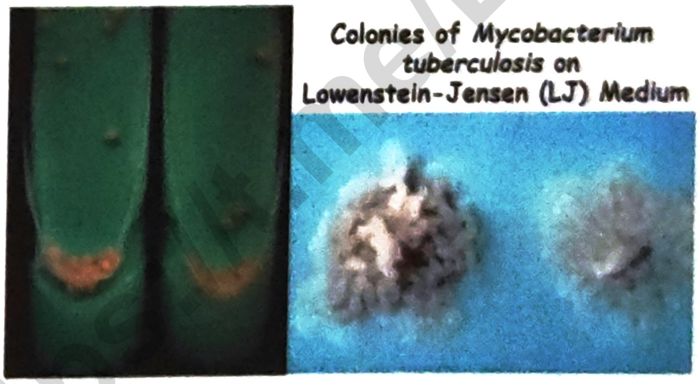
Main constituents: Egg Emulsion, Potato starch, Malachite green

Method for sterilization: Inspissation

Color of prepared medium: Bluish green colored opaque, smooth slant.

Intended uses:

- It is used for isolation of mycobacterium species.
- Colonies appear: dry, wrinkled, rough, buff, tough.



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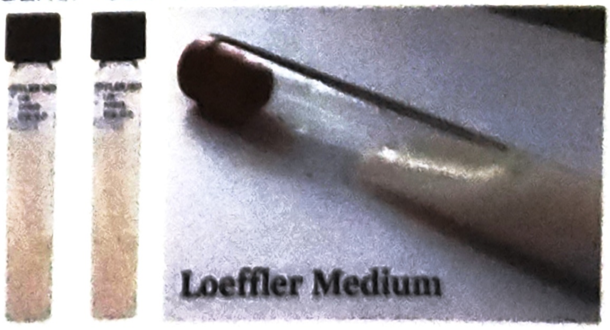
LOEFFLER SERUM SLOPE MEDIUM



PrepLadder

Media Type: Enriched medium

Main constituents: Serum, Egg, Nutrient broth

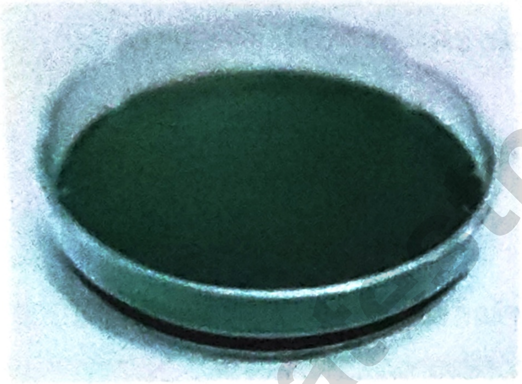
Method for sterilization: Inspissation

Color of prepared medium: Light yellow to brownish yellow

Intended Use:

- Early detection (6-8 hrs) of C. diphtheriae.
- Enhances production of metachromatic granules and appearance suggestive of Chinese-letter formation in methylene blue stain.
- Growth of cornyebacterium appear as minute, and cream colored colonies with slightly raised centers.

THIOSULFATE CITRATE BILE SALTS SUCROSE (TCBS) AGAR



Media Type: Selective medium

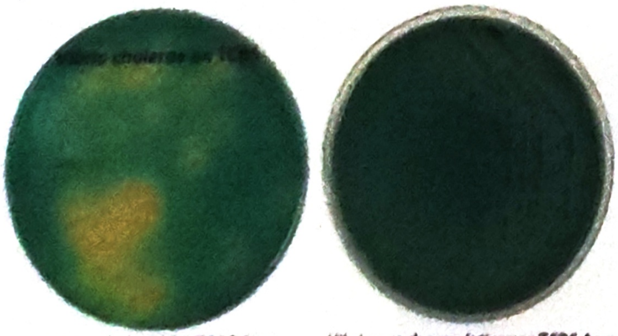
Main Constituents: Sucrose, Sodium citrate, Sodium thiosulphate, Bile salts.

Method for Sterilization: Autoclaving

Color of Prepared Medium: Bluish green colored clear to slightly opalescent gel forms

Intended Use:

V. cholerae produces large colored colonies after fermenting sucrose.



Vibrio cholerae on TCBS Agar *Vibrio parahaemolyticus on TCBS Agar*

- Non sucrose fermenter produces green colored colonies.

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POTASSIUM TELLURITE AGAR (PTA)

Media Type: Selective medium

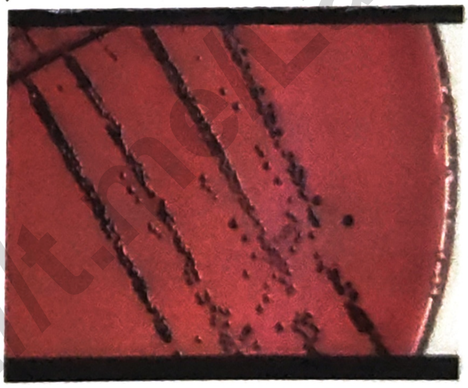
Main Constituents: Potassium Tellurite, Blood agar

Method for Sterilization: Autoclaving

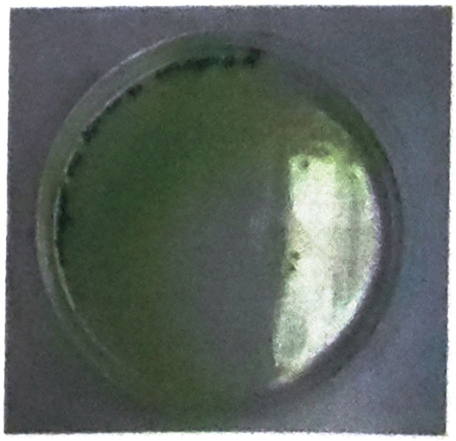
Color of Prepared Medium: Reddish brown colored, opaque gel form.

Intended use:

- It is used for isolation of C. diphtheriae
- C. diphtheriae produced black colored colonies on PTA



CETRIMIDE AGAR



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Media Type: Selective medium.

Main constituents: Cetrimide & Peptone

Magnesium chloride & potassium sulfate: Stimulates pyocyanin production

Method for sterilization: Autoclaving

Color of prepared medium: Cream to yellow

Indented use:

1. Isolation of P. aeruginosa
2. Demonstration of pigment

ROBERTSON'S COOKED MEAT (RCM) MEDIUM

Media Type: Anaerobic medium

Main Constituents: Beef heart meat particles: Contains reducing substances.

Unsaturated fatty acids: Creates low redox potential.

Hemin and Vitamin K: Enhance the growth of anaerobic bacteria.

Method for sterilization: Autoclaving

Color of prepared medium: Medium amber colored, clear to slightly opalescent supernatant over insoluble Brownish meat

Intended use:

- Used for isolation of Anaerobic bacteria
- It can be also used as Anaerobic transport medium

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Demonstration of Proteolytic and Saccharolytic activities:

Proteolytic bacteria: Blackening of meat particles. C. Tetani, C. botulinum A, B, F.

Saccharolytic Bacteria: Pinking of meat particles C. perfringens, C. difficle, C. botulinum C, D, E.

MUELLER HINTON AGAR (MHA)

Media Type: Antibiotic susceptibility testing medium.

Main Constituents: Acid hydrolysate of casein, Beef extract

Method for Sterilization: Autoclaving

Color of Prepared Medium: Cream to yellow

Intended use:

- To perform Antibiotic susceptibility testing (AST) against isolated pathogenic bacteria from clinical specimens.
- To perform MIC by E. test.
- MHA has been selected by the CLSI for several reasons:

- It is low in sulfonamide, trimethopirm and tetracycline inhibitors.

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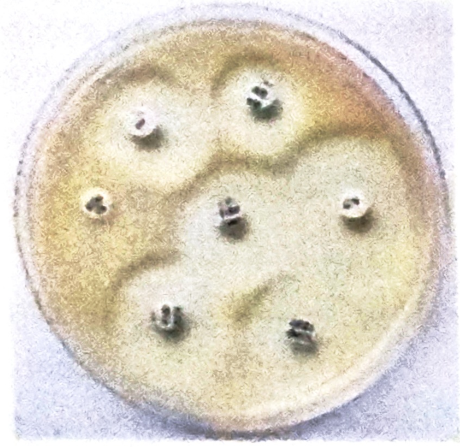
- It supports the growth of most non-fastidious bacterial pathogens



E-TEST

BRAIN HEART INFUSION (BHI) BROTH

Media Type: Enriched media

Main Constituents: Calf brain & Beef heart in infusion from, Peptone

Method for Sterilization: Autoclaving

Color of Prepared Medium: Sterile clear

Intended use:

- Used as blood culture media
- Used for anaerobic bacteria cultivation and the recovery of fastidious bacteria
- Useful for isolation and cultivation of fungi

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MIDDLEBROOK 7H9 BROTH

Media type: Special media for Mycobacterium

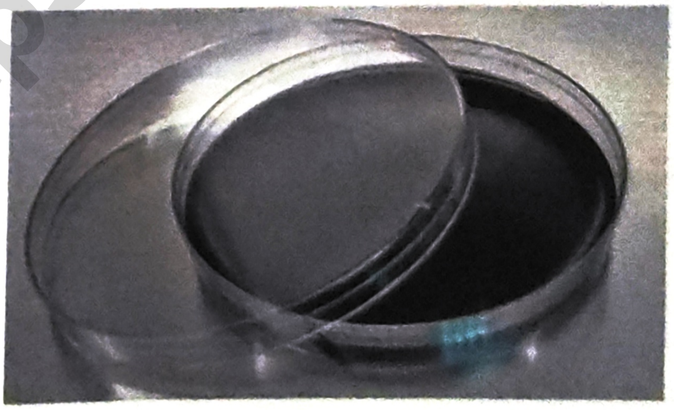
Method for sterilization: Autoclaving

Color of prepared medium: Light amber colored clear solution in tubes

Intended use:

- Middlebrook 7H9 Broth Base with added enrichment is recommended for cultivation and sensitivity testing of Mycobacterium tuberculosis
- Middlebrook 7H9 Broth with ADC (Albumin, Dextrose, Catalase enzyme) Enrichment used in MGIT system to recover Mycobacterium tuberculosis from clinical specimens.

BUFFERED CHARCOAL YEAST EXTRACT (BCYE) AGAR



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Media Type: Selective medium

Main Constituents: Yeast extract, ACES [N-(2-Acetamido) -2 aminoethanesulfonic acid] buffer, Charcoal.

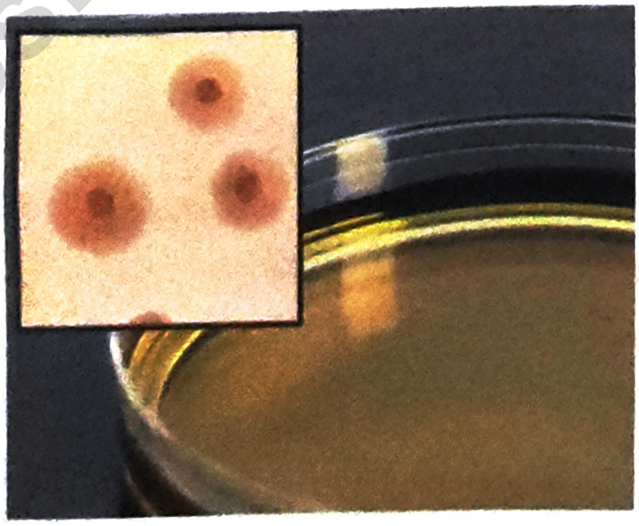
Method for sterilization: Autoclaving

Color of prepared medium: Grey-black colored to opalescent

Intended use:

- This medium is to be used for the isolation and presumptive identification of Legionella.
- It is a base medium used for the isolation of Legionella from clinical and environmental samples.
- Colony characteristics:
  - After 3-4 days of incubation, small white colonies of Legionella appear in BCYE agar which becomes grey in a few days
  - The isolate may show typical ground glass (or iridescent) colony appearance.
  - Colonies may be examined under UV light to reveal yellow pigment of BCYE of auto-fluorescence of Legionella colonies.

PLEURA-PNEUMONIA LIKE ORGANISM (PPLO) AGAR



Media Type: Selective Medium

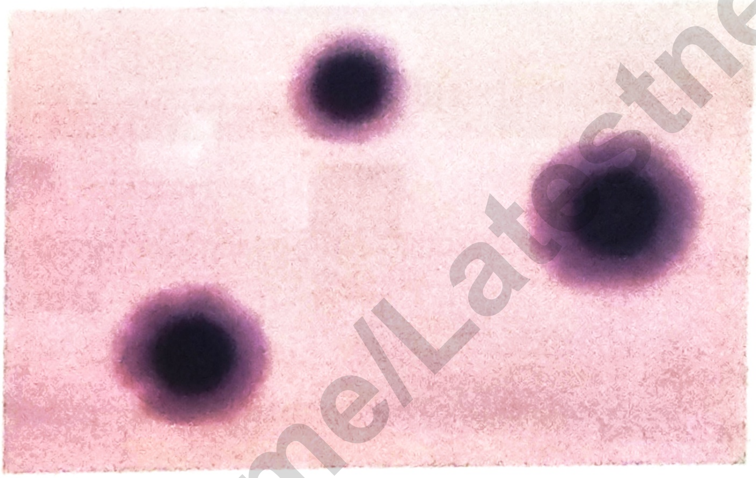
Main constituents: Yeast Extract, Horse Serum, Polymyxin B, Amphotericin B, Pencillin, Phenol Red

Method for sterilization: Autoclaving

Color of Prepared Medium: Yellow colored clear to slightly opalescent gel forms

Intended use:

- Recommended for isolation and cultivation of Mycoplasma species.
- Colony characteristics: Microscopic examination at 40-60X of inverted plates reveals that Mycoplasma are recognized by typical tiny "Fried Egg" colonies.



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SKIRROW'S MEDIUM

Media Type: Selective Medium

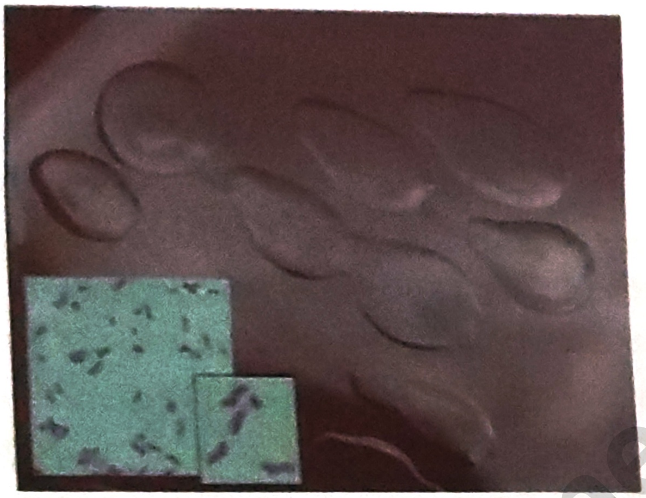
Main Constituents: Horse Blood, Casein Pancreatic Digest, Yeast Extract, Vancomycin, Trimethoprim, Polymyxin B.

Method for Sterilization: Autoclaving

Color of Prepared Medium: Reddish brown colored

Intended use:

- Selective media for the isolation of Campylobacter
- Produces effuse droplet like colonies
- C. →swarm on campylobacter media
- Positive oxidase test & Gram stain →gull wing-shaped Gram negative rods



Pike's medium

Media type: Transport medium

Main constituents: Casein, Tryptose, Yeast extract

Method for sterilization: Autoclaving

Color of prepared medium: Yellow colored clear solution without any precipitate

Intended use:

- Pike Broth is used for the selective enrichment and cultivation of streptococci from throat swabs and other clinical specimens.

AMIES MEDIUM

https://t.me/Latestneetpgnotes

PrepLadder

Media type: Transport media

Main constituents: Charcoal, Sodium thioglycollate

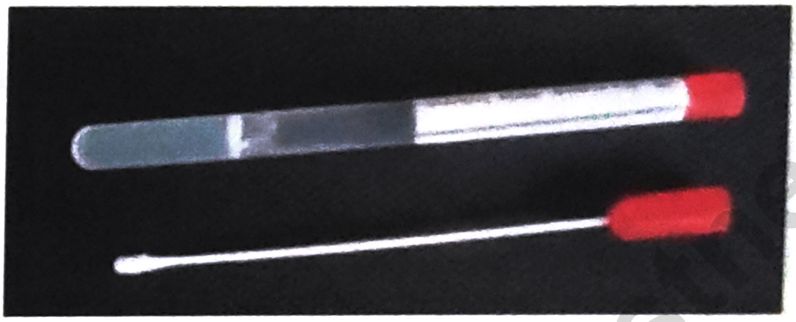
Method for sterilization: Autoclaving

Color of prepared medium: Black coloured opaque gel forms in tubes

Intended use:

Amies transport medium is used for collection, transport and preservation of microbiological specimens

STUART MEDIUM



Media Type: Transport medium

Main constituents: Sodium glycerophosphate, Sodium thioglycollate

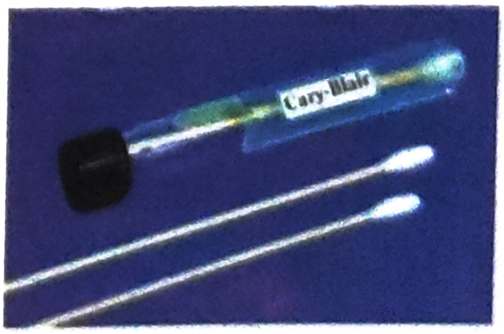
Method for sterilization: Autoclaving

Color of prepared medium: colorless to whitish colored

Intended use:

- Recommended for the preservation and transportation of Neisseria species.
- This medium is a chemically defined, semisolid, non-nutrient medium which prevent microbial proliferation. Because of this composition the medium ensures that microorganism present are able to survive for a sufficiently long period of time

CARY-BLAIR MEDIUM



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PrepLadder

Media Type: Transport medium

Main constituents: Disodium hydrogen phosphate, Sodium thioglycollate, Sodium chloride

Method for sterilization: Autoclaving

Color of prepared medium: Light amber coloured

VENKET RAMAKRISHNAN (VR) MEDIUM

Media Type: Transport medium

Main constituents: Crude sea salts, Peptone, pH-8.6

Method for sterilization: Autoclaving

Color of prepared medium: Transparent

Intended use:

- Transport medium for Vibrio

SABOURAUD DEXTROSE AGAR (SDA)

Media Type: Selective medium

Main constituents: Dextrose, Peptone, Low pH (5.6), Chloramphenicol

Method for sterilization: Autoclaving

Color of prepared medium: Light amber colored clear to slightly opalescent gel forms

Intended use:

Isolation and identification of fungi

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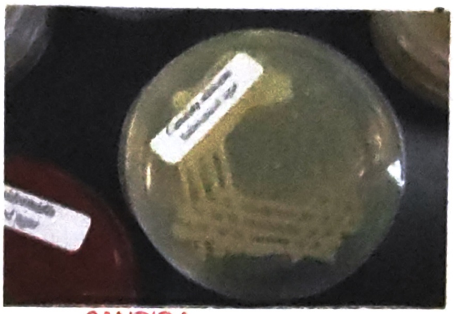
PrepLadder



CANDIDA

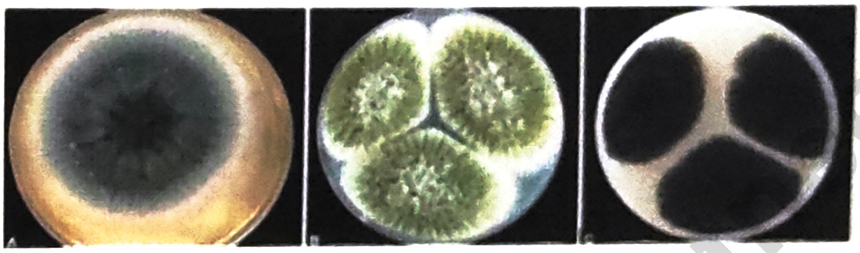
CRYPTOCOCCUS

- Aspergillus colonies on SDA:

  A. A. fumigatus

  B. A. flavus

  C. A. niger



TYPES OF BACTERIA

01:47:17

Bacteria	Alternate name
Bordetella	Bordet Gengou bacillus
Clostridium tetani	Nicolair's bacillus
Corynebacterium diphtheria	Klebs-Loeffler's bacillus Q
Corynebacterium pseudotuberculosis	Preisz-Nocard bacillus Q
Haemophilus aegypticus	Koch-Weeks bacillus
Haemophilus influenzae	Pfeiffer's bacillus Q
Klebsiella pneumonia	Friedlander's bacillus Q
Klebsiella ozaenae	Abel's bacillus
Klebsiella Rhinoscleromatosis	Frisch's bacillus Q
Mycobacterium tuberculosis	Koch's bacillus
Mycobacterium intracellulare	Battey's bacillus
Mycobacterium paratuberculosis	Johne's bacillus Q

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PrepLadder

GENETIC MATERIAL CODING FOR DIFFERENT TOXINS

- Plasmid borne (TPHA)
  - Tetanospasmin of C. tetani
  - Pertussis toxin of B. pertussis
  - Heat-labile toxin (LT) of ETEC
  - Anthrax toxin of B. anthracis
- Chromosomal (PPE Se Covered)
  - Cholera toxin (CT) of V. cholera
  - Shiga toxin of S. dysenteriae
  - Pyrogenic toxin B of S. pyogenes
  - Pertussis toxin of B. pertussis
  - Exotoxin A of P. aeruginosa
- Phage encoded (Bottle vich drinks paa)
  - Botulinum toxin of C. botulinum
  - Verotoxin (Shiga-like toxin) of EHEC
  - Diphtheria toxin of C. diphtheriae

INTRACELLULAR BACTERIA 01:53:10

Yersinia
Rickettsiae
Mycobacterium sps
Brucella, Bordetella
Chlamydia
Salmonella
Meningococcus
Legionella
Donovani granulomatis

Memory aid- Yaar Ritesh merekoe Bru Coffee Sale main La De

OBLIGATE INTRACELLULAR BACTERIA

M. Leprae
Rickettsiae
Chlamydia
Coxiella burnetti
T. pallidum

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PrepLadder

NONSPORING ANAEROBES 01:56:02

- Gram positive (Engineers loe MBA Program)
  - Eubacterium
  - Lactobacillus
  - Mobiluncus
  - Bifiodobacterium
  - Actinomyces
  - Propionibacterium

- Gram negative (BPL fuse)
  - B-Bacteroids
  - P-Prevotella, prophyromonas
  - L-Leptotrichia
  - F-Fusobacteriums

OXIDASE POSITIVE ORGANISMS
Memory aid (AL VL have PNH)

Aeromonas
Legionella micadedi
Vibrio cholerae
C. jejuni
Pseudomonas
Neisseria meningitides and gonorrhea
Haemophilus influenzae

Oxidase reagent is Tetra methyl para phenylene diamine dihydrochloride Q

CATALASE POSITIVE BACTERIA [MY LIPS are (R) SEALED]

M – Meningitidis Neisseria
Y – Yersinia
L – Legionella, Listeria
I – Influenzae H.
P – Pseudomonas, Pasturella
S – StaphQ
are (R) - HelicobateR
S – Shigella (except dysentriae - 1) Q
E (resembles C) – Coliforms
A – Atypical Mycobacteria
L – Listeria
ed

Others:
H. pylori
L. monocytogenes
Legionella
Nocardia
Brucella

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PrepLadder

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Or click this page to join

Organism and Their Infective Doses 02:04:35

ORGANISM	INFECTIVE DOSE
ETEC (Entero Toxic E. Coli)	$10^6 - 10^{10}$
EIEC (Entero Invasive E. Coli)	$10^8 - 10^{10}$
EHEC (Entero Haemorrhagic E. Coli)	10 – 100
Shigella	10 – 100
Salmonella	$10^3 - 10^6$
C. Jejuni	10^4
Y. enterocolitica	$10^8 - 10^9$
Vibrio	10^{10} (water source) $10^2 - 10^4$ (food source)

PLEOMORPHIC ORGANISM (MCH Very Very Progressive)
Mycoplasma Clostridium H. influenzae V. cholerae V. parahemolyticus Proteus

CLASSIFICATION

Based on O_2 Requirement 02:11:27

1. Obligate Aerobes
   - Needs desperate O_2
   - MTB
     Pseudomonas
     Brucella
     Bacillus
2. Obligate Anaerobes
   - Does not need O_2 at all
   - A → Actinomycetes
     B → Bacteroids
     C → Clostridium species
3. Facultative anaerobes
   - Changed anaerobe
   - Aerobe + anaerobe
   - Most of the medically important bacteria
   - Eg. Staphylococcus, streptococcus species

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PrepLadder

4. Microaerophiles
  - Need 5% O_2
  - Campylobacter
    Helicobacter

5. Capnophiles
  - CO_2 loving
  - Brucella abortus
    Pneumococcus
    H. Influenzae

Based on Temperature Requirement 02:19:56

P → Psyhcrophile → -10° C to +10° C

M → Mesophile → 20° C to 40° C [medically important bacteria]

T → Thermophile → › 45° C, › 55° C, › 60° C → (Better Option)

→ › 100° C [can manage]

→ Thermus aquaticus [typical example]

→ Used in PCR

→ Father of PCR → Karry B Mullis

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PrepLadder

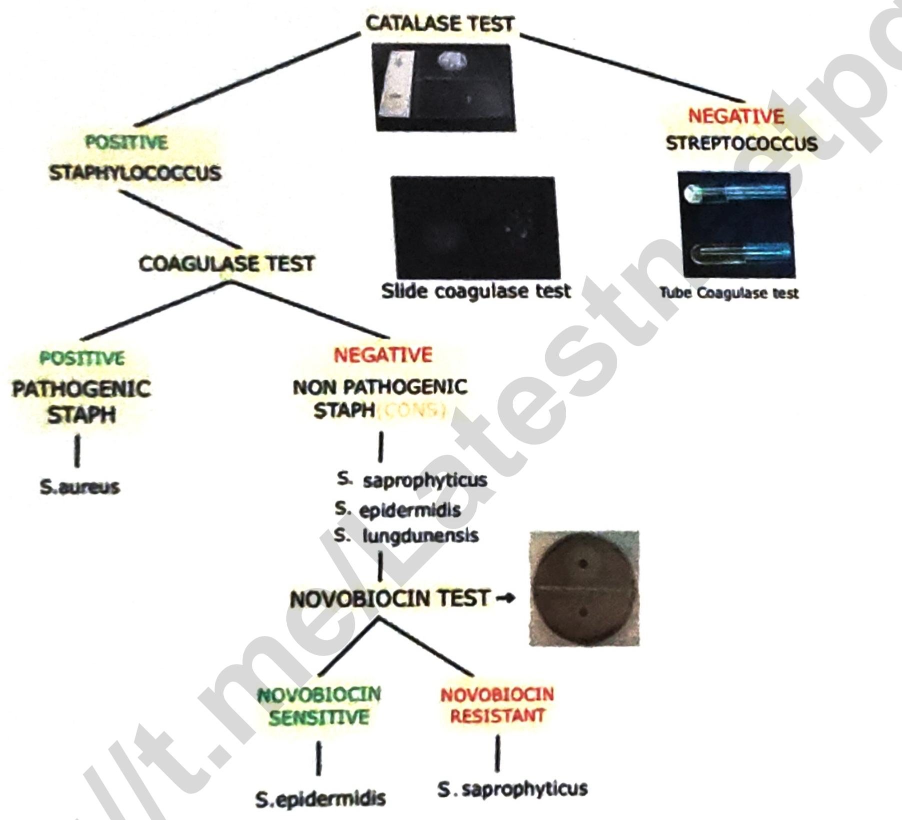
Unit: 2 Systemic Bacteriology

Chapter 2

GRAM +VE & GRAM -VE COCCI

GRAM POSITIVE COCCI

00:00:14



- Novobiocin test → done for Antibiotic testing
- Slide coagulase test → to determine bound form
- Tube coagulase test → to determine free form
- 3% H_2O_2 + Staph. colonies → Bubbles (Catalase Positive)

AST [Antibiotic Sensitivity Testing]

- Eg: novobiocin sensitivity test
- KBDDT method

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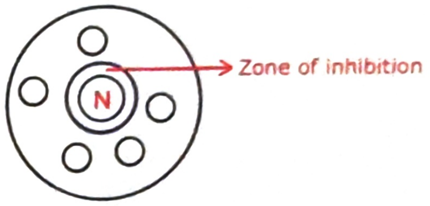
PrepLadder

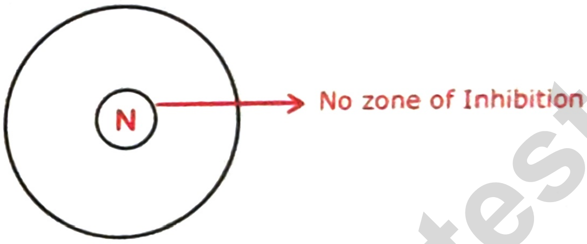
- Done on MHA [Muller Hinton Agar]
  NA [Nutrient Agar]
- 6 antibiotic discs can be put in an average petridish
- Kirby Bauer Disc Diffusion Testing Done

– Interpretation

Zone of Inhibition → Sensitive [Staph. Epidermidis]



No Zone of Inhibition → Resistant [Staph. Saprophyticus]



Coagulase Negative Staph (CONS) 00:07:18

STAPH EPIDERMIDIS 00:07:59

- Ⓝ Commensal of skin Epidermidis
- a/w INTERVENTIONS
  - IV canula, CSF Shunts, prosthetic valve surgery, catheters
  - Causes early prosthetic valve endocarditis [< 1yr]
- BIOFILM formation seen → helps in adherence,
  Antibiotic Resistance
- Novobiocin sensitive → zone of inhibition is present

STAPH SAPROPHYTICUS 00:11:44

- Young female develops UTI MC GPC involved - S. saprophyticus
- Young female develops UTI MC cause overall - E. Coli
- Novobiocin resistance - no zone of inhibition

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PrepLadder

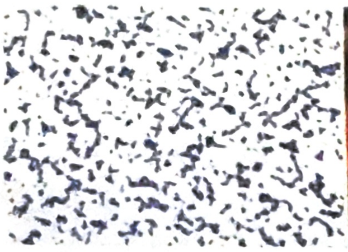
MC organisms/group involved in Early Prosthetic Valve Endocarditis → CONS

Single most cause for Early Prosthetic Valve Endocarditis → S. aureus

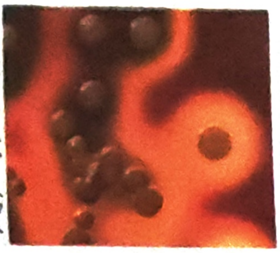
PATHOGENIC STAPH

STAPH AUREUS

00:15:25



S. aureus

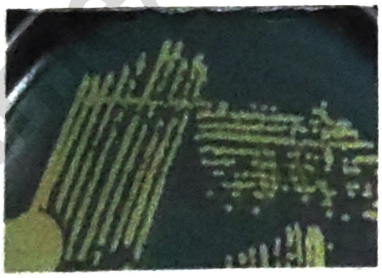


BLOOD AGAR

α, β, γ HEMOLYSIS

- Gram positive cocci
- Catalase +ve
- Coagulase +ve
- Gram staining → Grape like clusters (purple to violet)
- CULTURE MEDIA

1) NA [Nutrient Agar] → Golden yellow pigmentation
↓
Enhances on milk agar



Golden Yellow Colonies on NA

2) BA [Blood Agar]
- causes β hemolysis → Complete zone of hemolysis
- pin head size colonies

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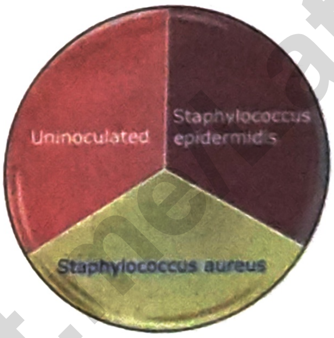
HEMOLYSIS

1. α Hemolysis 00:24:29
   - Partial hemolysis around colonies
   - Greenish discoloration is present
   - Unlysed RBC's can be seen under microscope
   - Eg → Streptococcus viridans, Streptococcus pneumoniae

2. β Hemolysis
   - Well defined clear colorless zone of Hemolysis around colonies
   - Eg → Streptococcus pyogenes

3. γ Hemolysis 00:26:30
   - No hemolysis around colonies
   - Eg → Enterococcus

3) MANNITOL SALT AGAR [Best Selective Media]



MANNITOL SALT AGAR

- In presence of Staph. aureus the agar changes/ transforms from pink to yellow

4) LUDLAM'S MEDIA [2nd best media]

5) SALT MILK AGAR

VIRULENCE FACTORS

1) Thickness of peptidoglycan layer
2) Teichoic acid
3) Clumping factor
4) Protein A

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PrepLadder

- Is present in COWAN-1 strain of Staph. aureus
- plays a role in co-agglutination
- has a property of binding with the Fc portion of Ig G

HEMOLYTIC FACTORS

5) Hemolysins ($\alpha, \beta, \gamma, \delta$)
- α Hemolysins
  - shows paradoxical action
- Inactivities at 70° C
- Reactivities at 100° C

- β Hemolysins / sphingomyelins
  - shows HOT & COLD PHENOMENON
- β Hemolysis starts at 37° C
- in S. aureus best seen at 4° C

- γ Hemolysin + PV Toxin [Panton Valentine Toxin] 00:27:52
  - Gives SYNERGOHYMENOTROPIC action [↑virulence of Staph. aureus]

6) Leucocidin [Panton Valentine Toxin]
- Kills leucocytes

7) Enzymes
- DNAase
- Thermonuclease
- Coagulase [plays the most important role in pathogenicity & virulence]
  - Tube coagulase → free form of coagulase
    → Better & more confirmatory
  - Slide coagulase → Bound form of coagulase
- Hyaluronidase
- Lipase
- Phosphatase

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PrepLadder

TOXIN MEDIATED ILLNESSES 00:30:28

1) Food Poisoning
   - S. aureus
   - IP → 1-6 hrs
   - Resembles Bacillus cerus emetic strain
   - S. Aureus → contaminated meat & milk products
   - B. Emetic → contaminated chinese fast food

   - d/t preformed toxin:
     - Enterotoxin
       - Enterotoxin A (MC)
       - Heat stable
       - Vagomimetic action /vagal stimulation

2) SSSS [Staphylococcal Skin Scalded Syndrome] 00:35:08
   - d/t Exfolliative / Enterotoxin, Toxins A & B
   - aka TEN → Toxin Epidermolytic Necrolysis in adults
     RITTER's disease in children

3) TSS [Toxic Shock Syndrome]
   - d/t TSS toxin [Enterotoxin F]
   - It is a super antigen [Non-MHC restricted]

↓

Bind laterally on T-cell receptors

↓

Activates > 20% lymphocytes

↓

Massive Lymphokine release

↓

Shock

- Rx: I.V fluids + Clindamycin
- d/t usage of contaminated vaginal tampons during menses

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PrepLadder

- Other Diseases a/w S. aureus — 00:40:40
  - i) Cellulitis
  - ii) Bullous Impetigo
  - iii) Carbuncles
  - iv) Furuncles
  - v) Acute osteomyelitis
  - vi) Acute mastitis
  - vii) Post op. parotitis
  - viii) Epidural / subdural abscess
  - ix) MCC of early PVE: } S. aureus
    MCC of native valve endocarditis } S. aureus
  - x) Iv drug abuser → Rt sided valve endocarditis
  - xi) Pnemonia → Pneumatocele
  - xii) Botryomycosis → Mycetoma like lesion
  - xiii) Tropical pyomyositis

MRSA [Methicillin Resistant Staph. Aureus] — 00:45:35

- DOC → vancomycin (a/w RED MAN SYNDROME)
- Mechanism of resistance

(1) PBP (Transpeptidase) → PBP 2a [main mechanism]
(PBP-Penicillin Binding Protein)
d/t MEC A gene aquisition;

MEC A GENE TYPES [6 TYPES]

I, II, III } Causes Nosocomial MRSA
↓
> 48 hrs of admission in hospital

IV, V, VI } Cause community Acquired MRSA

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PrepLadder

(2) Production of β Lactamases (Pencillinases)

LAB DIAGNOSIS

- Screening Tests

(1) CEFOXITIN → Resistant → MRSA (100%)

OXACILLIN → Resistant → MRSA (90%)

- Method: KBDDT
- Done on MHA [Muller-Hinton Agar]/ NA
- Add 4% NaCl
- Keep temperature of incubation → <37° C [30° -35°]

(2) ELISA for MEC A gene

(3) PCR for MEC A gene [best screening test for MRSA]

(4) Latex Agglutination Test for PBP2a

- Contact isolation done for MRSA
- Mupirocin ointment or Bacitracin ointment indicated for nasal carriers
- Best method to prevent nosocomical spread → Hand washing

TRANSDUCTION

- MC method of resistance transfer in S. aureus
- Bacteriophage mediated
- PHAGE TYPING can be done 00:58:26
- 23 types of phages are available
- 80/ 81A → MC phage (MCly responsible for outbreaks in hospital)

STREPTOCOCCUS 01:05:55

CLASSIFICATION

α Hemolysis	β Hemolysis (βHS)	γ Hemolysis
- S. pneumoniae - S. viridans	- Based on carbohydrate 'C' in the cell wall Classified into A-V [Lancefield Classification]	- Enterococcus - Faecalis - Faecium

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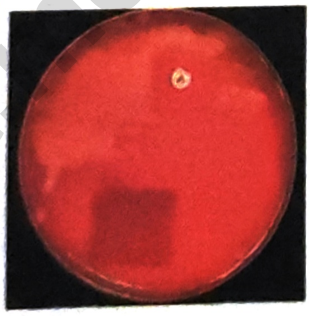
PrepLadder

	– A is divided into 80-100 groups based on 'M' protein [Griffith Typing] L - Lucknow C – City G – Good M – Morning – Group A [S. pyogenes] B [S. agalactiae] C [S. equisimilis] D [Enterococcus]	

	Group A Streptococci	Group B Streptococci
Bacitracin test	– Sensitive 	– Resistant Q
CAMP test	– Negative	– Positive
HHT	– Negative	– Positive
PYR Test	– Positive	– Negative

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PrepLadder

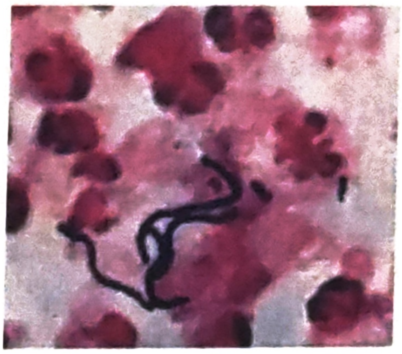
CAMP TEST → Christie Atkins Munch Peterson Test

HHT → Hippurate Hydrolysis Test

GROUP A STREPTOCOCCUS

Streptococcus Pyogenes

01:13:36



- Gram positive cocci
- Catalase -ve
- Arranged in violet colored chains on gram staining
  - Longest chain → S. salivarius

CULTURE MEDIA

1) CVBA [Crystal Violet Blood Agar]
   - Inhibits staph aureus
   - Produce β hemolysis
   - Pinpoint size colonies

2) Pike's media for S. pyogenes
   - Transport media

DISEASES

01:16:59

i) Cellulitis
ii) Impetigo (Honey colored crust)
iii) Eryiseplas [lymphatics involved]
iv) Pueperal sepsis
v) Necrotising fasciitis → flesh eating bacteria Q
vi) Acute pharyngitis
vii) Quinsy [sore throat]

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PrepLadder

viii) Scarlet fever toxin or Erythrogenic toxin A, B & C or Pyrogenic toxin → super antigen

↓

Activates > 20% lymphocytes

↓

SHOCK

ix) Non Suppurative Complications: 01:19:48

Acute Rheumatic Fever	Acute Glomerulonephritis
– H/O sore throat always – Any group A – Immune response marked – Complement levels are normal – Life long → prophylaxis of penicillin required – ASO Titres → > 200 Todd units	– H/O pyodermic infection / Sore throat – 49, 53-55, 59-61 – Mild to moderate immune response – Complement → ↓ ses, then normal – Sponteneous to come & spontaneous to go Anti DNAse B → > 300-350 units

GROUP B STREPTOCOCCUS 01:23:47

Streptococcus Agalectiae

- Normal commensal of birth canal
- MCC of Neonatal Meningitis
- Leads to Early or Late Neonatal Meningitis
- Early Neonatal meningitis:
  - 0-6 days
  - Prematurity / prolonged labour
  - Initially pneumonia → meningitis
  - High mortality (4.7%)
  - Serogroups : Ia, Ib, II, III, IV
- Late Neonatal meningitis:
  - 7-90 days
  - Less mortality (2.8%)

PrepLadder

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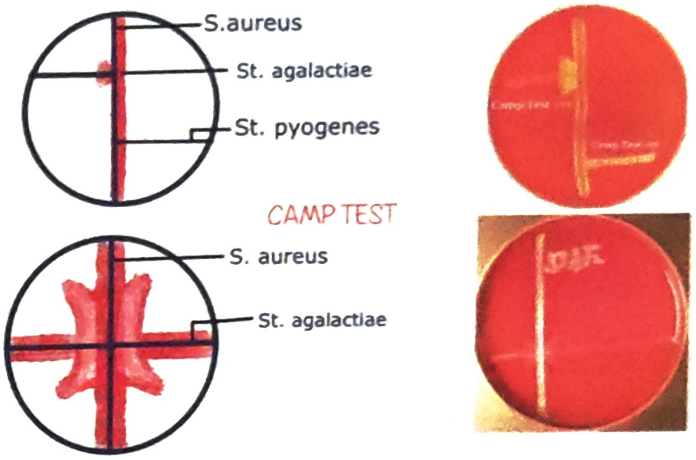
- Serogroups: Type III (MC)

CAMP TEST

01:28:30



Enhanced/ Butter Fly Zone of Hemolysis

- CAMP test +ve [enhanced/butterfly zone]
- HHT [Hippurate Hydrolysis Test] is +ve

GROUP D STREPTOCOCCUS

01:33:39

ENTEROCCUS

- E. Faecalis
  E. Faecium

NON ENTEROCOCCUS

- S. bovis
- S. gallolyticus
  ↓
  Colon cancer

- Resist
  - 6.5% NACL
  - PH = 9.4
  - 40% Bile
  - Temperature of > 60° C minutes for minutes
- Normal commensal of gut → 7-90 days
- Sx wound infection
- Lt. sided valve endocarditis
- DOC:
  - Pencillin + Aminoglycoside
  - Vancomycin [2nd line]

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PrepLadder

VANCOMYCIN RESISTANT ENTEROCOCCI

- With van gene aquisition [Van A-Van H]
- Van A is most resistant
- Doc → Linezolid

- Van gene → D-alanine-D-alamine
  D-alanine-D-lactate
  D-alanine-D-serine

CULTURE MEDIA

01:39:26

- Bile Aesculin Agar → Black Colored Colonies
- On Gram Staining →Spectacle like appearance is seen

ALPHA HEMOLYTICUS

STREPTOCOCCUS

- St. Viridans → leads to SABE, Late Onset Prosthetic Valve Endocarditis
- St. Mutans } Leads to Dental Caries
- St. Sanguis } Leads to Dental Caries

S. Viridians

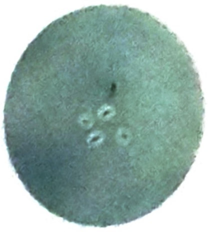
- Gram Positive Cocci

Biochemical Reaction

- B Bile Soluble } -ve
- I Inulin fermenter } -ve
- O Optochin sensitivity } -ve

STREPTOCOCCUS PNEUMONIAE [PNEUMOCOCCUS]

01:44:10



DIPLOCOCCI

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PrepLadder

- Gram postive Diplococci
- Lanceolate [Flame Shaped]
- Capsular polysaccharide
- Autolysin →Amidase enzyme
- Pneumolysin →membrane damaging toxin
- Carom coin appearance of colonies →DRAUGHT MAN APPEARANCE
- PSPA: Pneumococcal Surface Protein A
- IgA protease
- Pneumococcal surface protein C
- Adhesins

On the bases of capsular polysaccharide

- 90 types
- SSS typing Q

[Specific Soluble Substance Boxed Typing]

BIOCHEMICAL REACTION

- B Bile soluble
  I Inulin fermenter
  O Optochin sensitive
  } +ve
- Diseases

C →Conjunctivitis
O →Otitis Media (MC)
M →Meningitis [MCC of meningitis in adults]
P →Pneumonia
S →Sinusitis
Arthritis

- MCC of community Acquired Pneumonia (Lobar Pneumonia, Type 3 → most virulent)

VACCINES

1) 23 Valent Polysaccharide Vaccine
   - used for splenectomized individuals → as they are prone for capsulated organism infections

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PrepLadder

2) 13 Valent Conjugate Vaccine → Children

Indicated in:

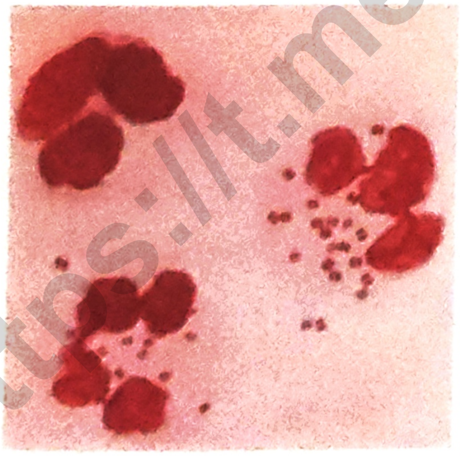
- for cochlear implant
- CSF shunt

- DOC for Pneumococcus: Penicillin

PBP → PBP_2b

GRAM NEGATIVE COCCI

01:58:57

MENINGOCOCCI	GONOCOCCI
– Capsulated	– Non capsulated o Some are capsulated
– Lens shaped	– Kidney shaped
– RCUT [Rapid carbohydrate utilization test] o Ferments Glucose & maltose	– RCUT [Rapid carbohydrate utilization o Ferments only Glucose
– Does not recruit Plasmid [For resistance]	– Recruits plasmid for resistance
– Extra/Intracellular	– Mainly intracellular
	
– Colony: Mostly circular – Natural habitat:	– Irregular margins

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PrepLadder

MENINGOCOCCI 02:05:51

- Gram –ve cocci
- Catalase +ve, oxidase +ve
- CAPSULATED
  - On the basis of capsule polysaccharides → 13 Serogroups
    - A, B, C, Y, W, E, X → most important Serogroups
    - A, B, C → most infective Serogroups
- IP: 3-4 days

- VIRULENCE FACTORS

1) Capsular polysaccharide
2) IgA_1 Protease
3) Lipo Oligo saccharide
4) OMP [outer membrane Proteins]
5) Pili

- 5-15% persons are normal nasopharyngeal carriers

↓

Spreads by Droplet infections

↓

Pulmonary involvement

↓

Haematogenous spread

↓

Meninges [Meningitis]
- Fever
- Vomiting (Projectile)
- Headache
- Rash present
- Neck rigidity → Kernig sign
  → Brudzinski sign

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PrepLadder

- LAB DIAGNOSIS

1) CSF EXAMINATION

- 3 samples taken
  - One for → Microscopy
  - One for → Pathology for cytology
  - One for → Biochemistry for Proteins & Sugars

2) CULTURE
   - MHA
   - THAYERMARTIN MEDIA (Modified Chocolate Agar)

3) GRAM STAINING SHOWS
   - Pink colored cocci, Lens shaped
   - Arranged extra/intra cellular

4) Blood Culture

5) Latex card agglutination test

6) PCR for meningococci (CSF/ Blood)

OTHER MANIFESTATION:

- Most dreadful complication → WATER HOUSE FRIDERICHSEN SYNDROME
  - B/L Adrenal Haemorrhagic necrosis
    +
    Purpuric Rashes
    +
    DIC
- In C_5- C_9 (MAC) complement deficiency → Recurrent Neisserial infections

- VACCINES 02:20:05

  MCV-4

  MENINGOCOCCAL CONJUGATE VACCINE
  - Active against A, C, T, W

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PrepLadder

- No good vaccine is available For group B
  - Group B capsule is Poorly immunogenic

- **TREATMENT**

- DOC For carriers → CIPROFLOXACIN
  2nd line → RIFAMPICIN
- DOC For cases → 3rd generation CEPHALOSPORINS
  [Note: If Pencillin given as option, it will be the answer]

GONOCOCCI

02:23:54

- Catalase & Oxidase +ve
- TYPING
  - Pili based typing done
  - AUXO typing [Nutrient based] done
- IP → 2-7 Days
- Causes Gonorrhea
- **VIRULENCE FACTORS**
  1) Capsular polysaccharide
  2) Ig A_1 protease
  3) Lipo Oligo saccharide
  4) OMP [outer membrane proteins]
  5) Opacity associated protein
  6) Pili
  7) Transferrin/ Lactoferrin

DISEASES

- GONOCOCCI

– In Male Acute Urethritis ↓ Purulent discharge (looks like semen) (flow of seeds) ↓ Later on Water Can Perineum	– In Female Cervicitis ↓ Mucopurulent discharge – Pregnant female ↓ New borns ↓ Ophthalmia neonatorum

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PrepLadder

- LAB DIAGNOSIS

1) SWAB

– Males	→	Urethral Swab
– Females	→	Endo cervical Swab
		High vaginal swab not taken
– Neonates	→	Eye Swab
Culture Media	→	THAYER MARTIN MEDIA
		Modified New york Agar
Gram Staining	→	Pink colored cocci, kidney shaped
		Arranged mostly intracellular
RCUT	→	Ferments only glucose
PCR	→	For gonococci

- COMPLICATIONS

02:33:24

1) WATER CAN PERINEUM
   - Occurs in males (urethritis)
   - Multiple fistula

2) FITZ HUGH CURTIS SYNDROME
   - Perihepatitis
   - Also a/w chalamydia

3) POLYARTHRITIS

↓

SUPPURATIVE ARTHRITIS

Deep Gonococcal infection (DGI)

- TREATMENT
  - Doc → 3RD gen CEPHALOSPORINS

  Plasmid recruit → ↑ Resistance against 3rd gen CEPHALOSPORINS

- OPHTHALMIA NEONATORUM IS MORE a/w CHALAMYDIA THAN GONOCOCCUS

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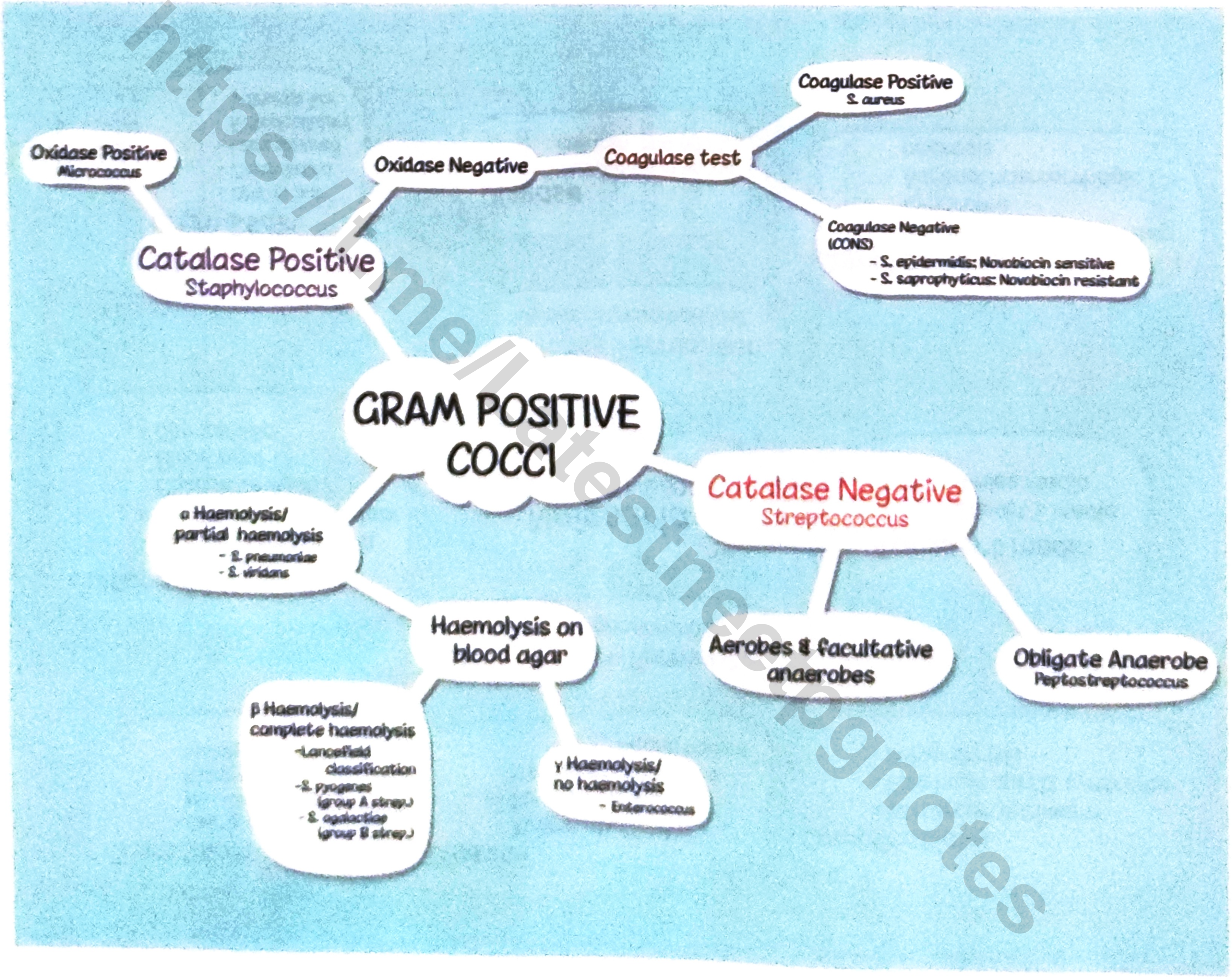

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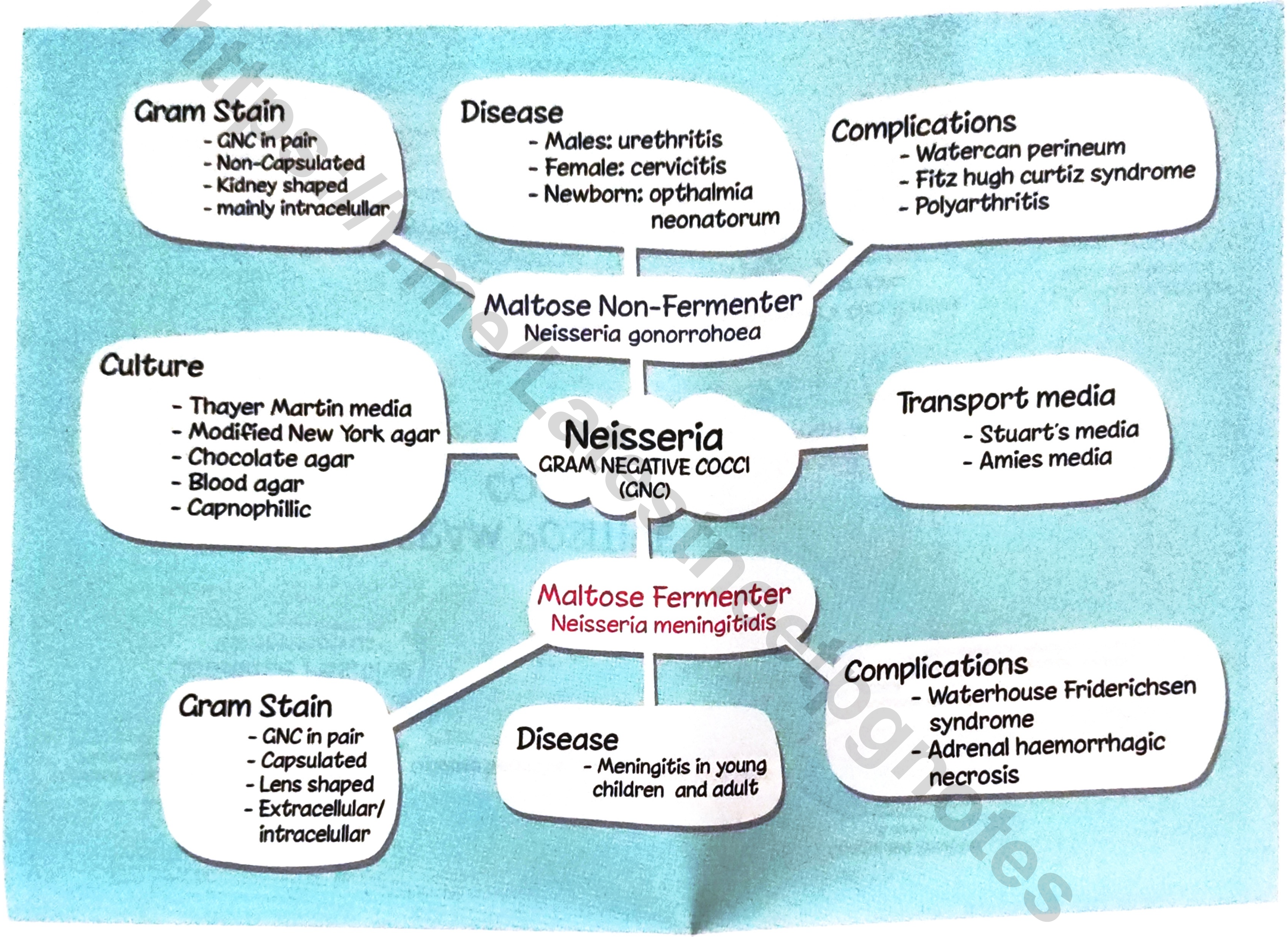
PrepLadder


Neisseria
GRAM NEGATIVE COCCI (GNC)
Maltose Non-Fermenter
Neisseria gonorrohoea
Gram Stain
- GNC in pair
- Non-Capsulated
- Kidney shaped
- mainly intracelullar
Disease
- Males: urethritis
- Female: cervicitis
- Newborn: opthalmia neonatorum
Complications
- Watercan perineum
- Fitz hugh curtiz syndrome
- Polyarthritis
Culture
- Thayer Martin media
- Modified New York agar
- Chocolate agar
- Blood agar
- Capnophillic
Transport media
- Stuart's media
- Amies media
Maltose Fermenter
Neisseria meningitidis
Gram Stain
- GNC in pair
- Capsulated
- Lens shaped
- Extracellular/ intracelullar
Disease
- Meningitis in young children and adult
Complications
- Waterhouse Friderichsen syndrome
- Adrenal haemorrhagic necrosis

Chapter 3

GRAM POSITIVE BACILLI

PART-1

CORYNEBACTERIUM DIPHTHERIA 00:00:19

(Kleb Loffler's Bacillus)

GRANULES

Metachromatic granules/ Polymetaphosphate granules/ Volutin Granules/ Babes ernest granules

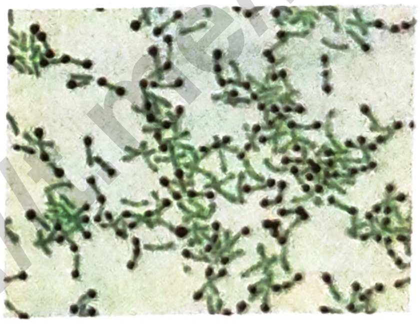
- These are Energy deposits
- Stained by

P → Ponders Stain

A → Alberts Stain

N → Neisser Stain

- Cuneiform arrangement/chinese letter pattern



Chinese Letter Pattern

BIOTYPES

		Colonies on PTA	Toxigenic strains	Virulence	Occurrence
1) Gravis	Daisy's	Daisy Head Appearance	100%	Severe	Epidemic
2) Intermedius	Frog eggs are	Frog Egg Appearance	95-99%	Moderate	Epidemic
3) Mitis	Poached	Poached Egg Appearance	82-85%	Mild	Endemic

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PrepLadder

DISEASE:

DIPHTHERIA

- Pseudo membrane (characterstic)
  (Greyish white membrane)
- MC site: Faucial
- Bleeds on touch
- Most dangerous → laryngeal
- Complication : Bull Neck → Neck edema
- Chronic complication: polyneuropathies of myocarditis

Diphtheria Pseudo Membrane

Bull Neck Edema

Lab Diagnosis

Throat swab

- Gram staining

Culture on:
- LSS (Loeffler's Serum Slope)
  ↓
  For early diagnosis (6-8 hrs)
- PTA → Definitive diagnosis best selective media

OTHER CULTURE MEDIA's

H → Hoyle's media
M → Macleod's media
T → Tinsdale media

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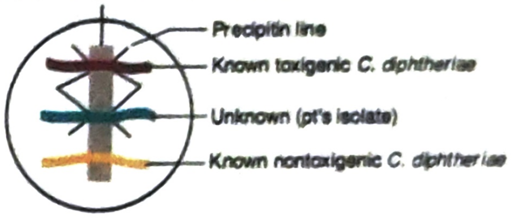
PrepLad

GS/AS →Cuneiform agents

TOXIN DEMONSTRATION

1) Elek Gel Precipitation Test
2) PCR For tox gene
3) ELISA For toxin Production



Elek Gel Precipitation Test

SCHICK TEST

Determines susceptibility of an individual to diphtheriae

If positive → needs immunization

If negative → already immunized

Neutralization test

- Diphtheria is TOXEMIA not Bacteremia

DIPTHERIA TOXIN

- Made of A+B [Bipartite toxin]
- Strong exotoxin
- Needs iron for it's action 0.1mg/dL
- Needs optimum Iron for it's action
  - A → ADP Ribosylation of EF - 2 + B (Helps in Binding)

↓

Inhibition of Protein Synthesis

↓

Cell Death

A B	} Components
C D	} C.diphtheriae ↓
E F	Acts on EF

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PrepLadder

- Every corynebacterium diphtheriae is not pathogenic, but made pathogenic by phage tox gene → LYSOGENIC CONVERSION

CUTANEOUS DIPHTHERIA 00:20:59

- Punched out ulcerative lesion
- Not toxin mediated

Rx:

- ADS
  - Anti Diphtheric Serum
  - In severe cases: 80,000-1,00,000 units
- Antibiotic:
  - Penicillin or Erythromycin
  - Better to give in initial stage of disease i.e., before toxin production

- For carriers: Erythromycin

- Protective Titers against Diphtheria
  - Antitoxin titer 0.01 unit/mL (Following vaccination)

NON CORYNEBACTERIUM DIPHTHERIAE 00:24:36

- C. parvum → immuno modulator
- C. jeikum → MDR isolate
- C. minutissimum → Causes Erythrasma
  → Coral red fluorescence is seen
- C. pseudo TB → Causes animal diseases
  → aka PREISZ NOCARD BACILLI
- C. urealyticum → Pyelonephritis
  → Struvite stones
- C. tenvis → Trichomycosis anillearis
  → Pigmented nodules in axillary hair

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PrepLadder

BACILLUS ANTHRACIS

00:29:11

- GPB [Gram Positive Bacilli]
- Non motile
- Capsulated
- Polypeptide
- Spore bearing (Box Car Appearance)
- certain size spores used in category A Bioterrorism
- Bioterrorist weapon → Category A

ANTHRAX TOXIN

- Made of TRIPARTITE TOXIN
  - E → Edema Factor
  - P → Protective Factor
  - L → Lethal Factor
- Acts Synergitically → ↑ CAMP

CAPSULE

Made of Polypeptide or Polyglutamate

MC FADYEAN'S REACTION

- Demonstrates capsule
- Culture growth + Loeffler's Methyle Blue → CAPSULAR STAINS

LAB DIAGNOSIS

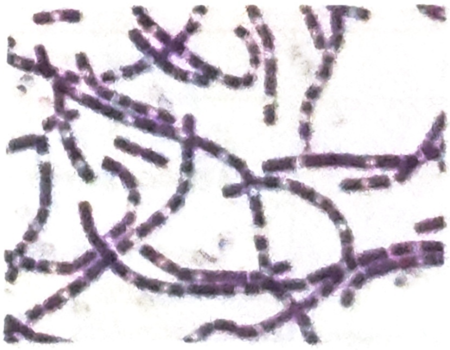
- Ascoli's Thermo Precipitation Test (ASCOLI → Anthrax antigen)
- PCR For Bacillus Anthracis

M → Medusa head appearance

I → Inverted fir tree appearance
→ seen in Gelatin Blue stab culture

S → String of pearl appearance
→ seen in Penicillin agar

Bamboo → Bamboo stick appearance



Bamboo Stick Appearance on Gram Staining

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PrepLadder

CULTURE MEDIA

PLET MEDIA [Best selective media]

P → Polymyxin

L → Lysozyme

E → EDTA

T → Thallous acetate

DISEASES

CUTANEOUS ANTHRAX [HIDE PORTER'S DISEASE]	PULMONARY ANTHRAX [WOOL SORTER'S DISEASE]

• Black Eschar → Edema
• Pustule with non pitting edema
  – Malignant Pustule
	• Occupational Hazard
	• Hemorrhagic Pneumonia
	• Causes Hemorrhagic Pneumonia, mediastinal hemorrhagic necrosis
	• Prevented by DUCKERING
	  Pre treat the wool by Formaldehyde (sporocidal)

INTESTINAL ANTHRAX

– Spread through eating contaminated wild animal meat ingestion

DOC: Ciprofloxacin & clindamycin x 60 days

BACILLUS CERUS

00:44:46

EMETIC STRAIN	DIARRHEAL STRAIN
– IP → 1 to 6 hrs – Causes Food poisoning ○ d/t chinese food (mimics S. aureus food poisoning) – In action it resembles Heat stable toxin of E. coli → ↑ CGMP	– IP → 8-16 hrs – Contaminated meat & milk – Resembles Heat labile toxin of E. coli → ↑ CAMP

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CULTURE MEDIA

- MTPA [Mannitol Yolksac Polymyxin Phenol Red Agar] MEDIA (MYPA is Cereus Fellow)

OBLIGATE ANAEROBES

CLOSTRIDIUM

00:53:11

- GPB
- Non capsulated
- Motile → STATELY [Slow Form of swarming]
  Exception → Cl. perfringes [capsulated, non motile]

CULTURE MEDIA

ROBERTSON COOKED MEAT MEDIA

Robertson Cooked Meat Medium

- If meat pieces turn PINK → Sacchrolytic
  ↓
  → Cl. perfringens
  → Cl. novy
  → Cl. septicum
  → Cl. botulinum C, D & E
- If meat pieces turn BLACK → Proteolytic
  → Cl. tetani
  → Cl. botulinum A, B & E

SPORE ARRANGEMENT (Bulding spore)

- Terminal & round → DRUMSTICK APPEARANCE → Cl. tetani
- Sub terminal spore → most clostridia
- central spore → Cl. bifermentans

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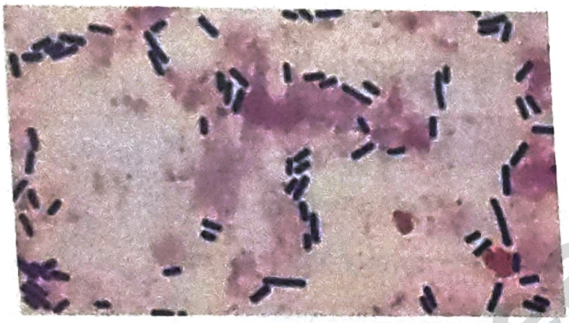
PrepLadder

METHODS OF ANAEROBIOSIS

- Mc intosh filde's jar
- Closed jar with candle (Obsolete)
- Gas pak (latest)
    - Chemical method of anaerobiosis
    - MC used

CLOSTRIDIUM PERFRINGENS/ WELCHII



Clostridium Perfringens

- 13 major & minor toxins are present
- α, β, epsilon, theta, iota
- Type A - E
- Most infections are caused by Type A; α toxin

DISEASE

1) Gas Gangrene/ Clostridial Myonecrosis 01:03:44

- IP → 1-2 days
- C/F → Pain
  Crepitus → Gas in the deep compartment
  (low oxidation reduction potential)
- Treatment
    - For Initial lesion
        - Surgical debridement & Antibiotics
    - For advanced case
        - Amputation of the limb & Antibiotics
- Gas Gangrene caused by
  P → Cl. Perfringens → Brick shaped
  N → Cl. Novyii

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PrepLadder

S → Cl. Septicum →Boat shaped CITRON bodies

- Gram staining:
  - Brick shaped
  - CITRON bodies
- Lab diagnosis
  - RCM →Sacchrolytic
  - PCR for Cl. perferingens

2) Food Poisoning 01:09:52

- Type A; toxin
- d/t contaminated meat products
- IP →8-24 hrs
- Involves Heat labile enterotoxin
  - But spores are Heat Resistant

3) Pigbel 01:11:25

- d/t uncooked/ contaminated pork consumption
- causes necrotizing enteritis
- d/t β toxin Type C

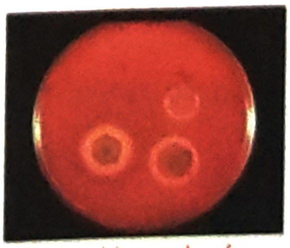
REACTIONS (Senior NTR)

(1) Stormy fermentation of litmus milk

(2) Nagler's reaction

(3) Target Hemolysis (Double zone of hemolysis)

(4) Reverse camp test



Target Zone Hemolysis

Stormy Fermentation

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PrepLadder

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1) NAGLER's reaction

- d/t α-Lecithinase or Phospholipase
- it is a neutralization reaction (toxin- Antitoxin reaction)
- done on Egg Yolk Agar

2) Stormy fermentation in litmus milk

- Immense fermentation of litmus milk

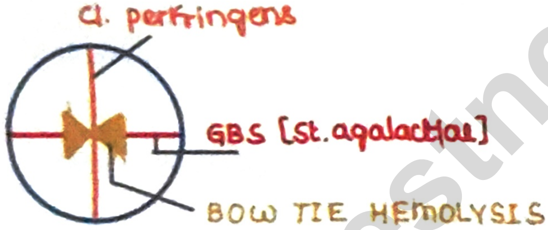
3) Target Hemolysis (Double zone of hemolysis)

- Inner zone → d/t θ toxin

Outer zone → d/t α toxin

4) Reverse CAMP test

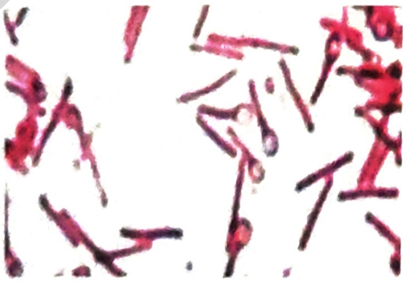
- Bow Tie Hemolysis with St. agalactiae



Reverse CAMP Reaction

CLOSTRIDIUM TETANI 01:20:02

- Tetanus (8th day disease)
- IP: 6-10 days
- If IP < 6 days → Grave prognosis


Drumstick Appearance of Cl. tetani

Toxins

1) Tetanolysin

- Hemolysis
- O_2 labile
- Resemble to toxin of S. pyogenes & S. pneumoniae

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PrepLadder

2) Tetanospasmin

- More dangerous
- Neurotoxin
- Causes presynaptic inhibition of Glycine/ GABA (inhibitory neuro- transmitters)

  ↓
- Constant stimulation

  ↓

  Lock Jaw (Trismus)

  ↓

  Opisthotonus Posture (Bad prognosis)

  ↓

  ↑ Deep tendon reflexes

Neonatal Tetanus

- India is free of neonatal tetanus d/t
  - Clean practices
  - Antenatal immunization
  - Hospital deliveries

Lab Diagnosis:

- Excised tissue fragment + RCM → Meat pieces turn Black
- PCR for Cl. tetani
- Mouse toxigenicity testing (Slightly proteolytic)

PREVENTION OF TETANUS AFTER INJURY

	SIMPLE WOUND	OTHER WOUND
Category A	Nothing required	Nothing required
Category B	Toxoid 1 dose	Toxoid 1 dose
Category C	Toxoid 1 dose	Toxoid 1 dose + HT Ig
Category D	Complete toxoid dose	Complete toxoid dose + HT Ig

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Category A: TT/Booster in last 5 yrs
Category B: Complete course in last 5-10 yrs
Category C: Complete course beyond 10 yrs
Category D: Unknown status

DOC for Tetanus: Metronidazole

Antitoxin Titres: > 0.01 units/mL

CLOSTRIDIUM BOTULINUM

Toxins

1) Neurotoxins (main) → most potent toxin on earth
2) C_2 (Enterotoxin) → a/w canned food poisoning

3) Botulinum Toxin:

- Zn dependent protein
  - Heavy Chain
  - Light Chain
- Anticholinergic action → Flaccid paralysis
- Acts on:
  - Most common nerve terminals
  - NMJ, Post ganglionic parasympathetic nerve ending
  - Peripheral ganglia
- BT binds to Ach receptor → Blocks release of Ach
  ↓
  Flaccid Paralysis
- Used in: Strabismus, Blepharospasm, Myoclonus
- BT can be produce by:
  - B → Cl. Butyricum
  - B → Cl. Baratti
  - A → Cl. Argentimense

INFANT BOTULISM

01:40:02

- a/w Sporulated honey ingestion
- Clinical Features
  - Flaccid paralysis → Floppy Infant Syndrome

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Lab Diagnosis

- RCM → Meat pieces → Pink
  → Black
- PCR for Cl. Botulinum
- Cultured on blood agar under anaerobic conditions

Neurotoxin Uses

1) Botox treatment → to remove wrinkles
2) Eye Surgeries
   - Blepharospasm
   - Strabismus

Diagnosis

1) PCR for Cl. botulinum
2) Culture for Cl. botulinum

CANNED FOOD POISONING

Clinical features

- Dysphagia
- Diplopia
- Disarythria
- Constipation (d/t ↓Ach)
- ↓DTR

CLOSTRIDIUM DIFFICLE

01:42:28

- Difficult to isolate
- Clostridium Difficle Associated Diarrhea (CDAD)
  - If not Rx in time, leads to Pseudomembranous colitis
  - Normal commensals of gut (Cl. difficle)
  - a/w long term use of 3rd generation → cephalosporin, clindamycin

Normal flora destroys

↓

Cl. difficles attaches to gut

↓

Bloody Diarhoea

Ulcer formation

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PrepLadder

* DOC: Oral Vancomycin

Lab diagnosis

1) Toxin Demonstration in stool [Entero toxin (A) + Cyto toxin (B)]
   Demonstrated on Hep 2 cell lines (commonly used for viral isolation)
2) PCR for tox gene
3) ELISA for toxin production
4) Sigmoidoscopy/ Colonoscopy to visualize any ulcer
5) Culture on CCFA (Cefoxitien Cycloserine Fructose Agar)

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PrepLadder

Corynebacterium diphtheriae
(GRAM POSITIVE BACILLI)

Microbiology

Culture Media

- LSS (Loeffler serum slope)
  - Shows growth in 6-8 hr.
- **Potassium tellurite agar :** best selective media

Staining

P → Ponder's stain

A → Albert's stain

N → Neisser's stain

- Metachromatic/ Babes Earnst/ Volutin granules
- Cuneiform/ Chinese letter pattern

Albert's stain

Clinical features

- **Faucial pseudomembrane**
  - Tough, grey, leathery
- **Bull's neck :** Cervical lymphadenopathy

Toxin

- Exotoxin
- Bipartite (A & B subunits)
- MOA : Protein synthesis inhibition in cell wall
- Lysogenic conversion
- β phage (tox+) gene.

Toxin Detection

- Elek's gel precipitation test
- PCR for tox gene

Complications

- Myocarditis
- Polyneuropathies

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PrepLadder

Chapter 4

GRAM POSITIVE BACILLI

PART - 2

MYCOBACTERIUM TUBERCULOSIS

00:00:14

- 20% Acid fast
  - Ability to resist decolorization
  - Depends on Mycolic Acid content

- Virulence Factors

1) Cord factor
2) Lipo arabinomannan
   - helps in attachment
   - LAM Ag detected in urine

These 2, prevent phagolysosomal fusion

3) HSP (Heat Shock Protein)

Pathogenesis

1) Human (Pulmonary TB)
2) Milk source

Mode of Transmission

- Inhalational mode
  - By droplet nuclei
  - Generated during coughing & sneezing

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Prep

SEQUENCE OF PATHOLOGICAL EVENTS

- Droplet nuclei (Tubercle bacilli) are inhaled

↓

Adhesion to macrophages

Mycobacterial Surface

(LAM)

↓

Binds to complement Receptor & Mannose receptor

(Present are macrophages)

↓

Internalization of bacilli

- Phagocytosis
  - C_3b mediated enhancement (opsonization of bacilli)
- Survival Inside The Macrophages
  - LAM
  - Cord factor

  } Prevent phagolysosomal fusion
- Macrophages Rupture

↓

Release Bacillary content which infect other phagocytes & cycle continues

HOST IMMUNE RESPONSE

00:10:47

- Macrophages present

MTB Ag to T helper cells (TH cell)

TH_1 | TH_2

TH_1

↓

IL_2

IFN γ

↓

Activates monocytes &

macrophages further TH_2

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PrepLadder

- Activation of TH_1 cells
  - Macrophage activation response (MAR)
  - Tissue damage response

Macrophage Activation Response

- Majority → Show resistance to infection (able to contain bacilli)
- INF activates resting alveolar macrophages

  ↓

  Killing & digesting of Tubercle bacilli starts
- Activated macrophages aggregate around centre of lesions & forms GRANULOMA

Hard Tubercle

- Hard (initial stage)

  Central zone (activated macrophages & Epithelioid gaint cells)

  Peripheral zone of lymphocytes & fibroblasts

Soft Tubercle

- Central part undergoes "Caseous necrosis" (Cheese like Necrotic material)

TISSUE DAMAGING RESPONSE 00:17:19

- Happens in minority
- MAR is weak → bacilli is more virulent

  ↓

  Immense delayed Hypersensitivity

  ↓

  Lung destruction

- Caseous necrosis 00:18:29

  ↓

  Liquifies

  ↓

  Large no of bacilli

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↓

1) Directly drain into the airways
2) Lymphatic spread
3) Hematogenous spread

PRIMARY PULMONARY TB 00:27:30

- Exogenous infection with tubercle bacilli
- Age: Children
- Sub pleural (upper part of lower lobe or lower part of upper lobe)
- Ghon's focus → Fibrotic nodular lesion
- Ghon's focus + Hilar LAP → Ghon's complex (Primary Complex)
- Calcified Hilar LAP → Ranke's complex

POST PRIMARY / SECONDARY PULMONARY TB

- Due to exogenous re-infection
- Reactivation of primary lesion
- Presence of calcified nodules → Assman's focus
- Mostly infraclavicular

- Hemotogenous spread
  - In apex of lung
  - Simon's focus
  - Involves apical or posterior segments of upper lobe

- Lesions undergo necrosis

  ↓

  Tissue destruction
  (Cavity formation)

Extra Pulmonary Involvement 00:34:35

- MC extra pulmonary involvement
  L.N > Pleural > Genito Urinary Tract
- Skin TB
  MC primary lesion → Lupus vulgaris 'Apple Jelly Nodules'
  2nd MC lesion → Scrofuloderma

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PrepLadder

- MC site for GI TB → Ileo caecal junction
- Skeletal TB → Pott's spine
  → Paravertebral cold abscess
- CNS TB
- Can Manifest as: i) TB meningitis
  ii) Tuberculoma (MCICSOL)

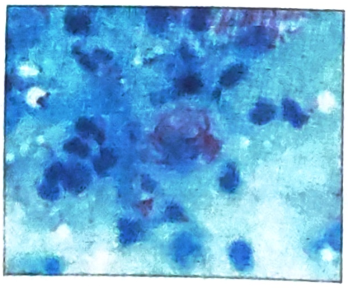
LAB Diagnosis

1) Sputum Examination

- 2 sputum samples taken
  (i) Onspot sample
  (ii) Early morning sample

- Concentration method → Petroff's method
- Thick sputum + NaOH/Hcl → Liquified sputum
- For Bacilli to be visible on:
  - ZN smear → 10,000 bacilli/mL of sputum
  - Culture media 10-100 bacilli/mL of sputum

a) ZN STAINING



Acid fast staining
ZN staining

ZN STAINING

↓

Carbol fuschin

↓

Intermittent heating

↓

20% H_2SO_4 (Decolouriser)

↓

Methylene Blue

↓

Blue background; Pinkish to red colored AF Bacilli

- (Modification of acid fast stain)
- (Kinyoun method of staining)
- Cold method of staining more amount of carbol fuschin is used

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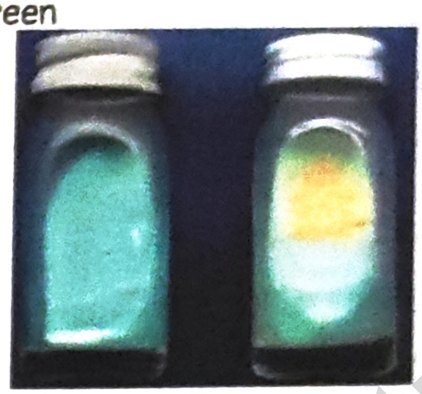
Prep

here & phenol concentration is high → Duration is ↑ed

b) Culture on LJ Media (Lowen Stein Jensen Media)

- Eggs are used for solidification
- Selective media agent → Malachite green
- Green colored media
- Rough, Tough, Buff Colonies
  - Rough → Irregular colonies
  - Tough → Difficult to remove
  - Buff → Yellowish Brown
- Takes 6-8 wks



LJ Media

2) Auramine – Rhodamine Staining

- Florescent staining
- Done in cases of more load
- Drawback → false positive
  Rates are high

00:49:49

00:51:09

3) Automated liquid culture

- BAC TEC/ BACT – alert
- Use liquid broth → Middle brook 7H9

BACTEC MGIT

Principle of BAC TEC: O_2 sensitive fluorescent compound in broth

- Initial Phase: O_2 is more
  Fluorescence is quenched
  In MTB: O_2 is less
  Fluorescence is more

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Molecular Methods 00:54:45

- PCR based → Target on IS6110 gene
- Turn around time 2 hrs
- Can't differentiate between live/ Active/ Latent TB
- CBNAAT (Gene xpert Rif)
  - Cartridge based Nucleic Acid Amplification Test
  - Rifampicin drug resistance is also detected (based on rPOB gene)
- LPA → Line Probe Assays
  → PCR based + MDR detection
- MGIT → Mycobacterium Growth Indicated Tube
  → Can be used for drug resistance

Tests Not Done In India 00:59:56

- Serological Test: Banned in India → No ELISA & no card test
- MTP 64 Ag detection → Immunochromatography test
- Biochemical test → NIACIN

DIAGNOSIS OF LATENT TB

Quantiferon TB Gold Assay

(IGRA- Interferon Gamma Release Assay)

Mycobacterial Antigens

↓

Stimulate

Blood sensitized T lymphocytes

↓

Measure Interferon activity

(Activity of sensitized T lymphocytes)

↓

Prior exposure will be known

Tuberculin Test / Montoux Test 01:08:06

- 0.1 ml PPD (Purified Protein Derivative)
- Into flexor aspect of forearm

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PrepL

- After 3 days, measure Induration (Hardness Diameter)

  <5mm → Negative

  5 - 10 mm → Equivocal

  >10 mm → +ve
- Erythema is not to be measured
- False Positive
  - Recent BCG vaccination
  - Atypical mycobacterial infection
- False negative (IN THOSE WITH IMPAIRED CMI)
  - HIV +ve (Advanced Stage)
  - Miliary TB
  - Malignancy
  - Immunosupression
- Delayed hypersensitivity reaction

BCG (Bacillus Calmette Guerin) Vaccine

- Derived from Danish 1331 strain of Mycobacterium bovis
- Intra dermal
- Diluents →Normal Saline
- Efficacy →0 - 80%

- TB is a Notifiable Disease
- Serological tests (MTS card tests, MTB Elisa) are banned in India

MDR TB 01:11:54

- Resistance against H & R
- Resistance against H R Z E S
- But, resistance against Z E S not MDR TB
- For MDR TB Resistance against H & R should be there

XDR TB

Resistance against 1st line drugs (HRZES)

+

One of the quinolones in 2nd line drugs

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PrepLadder

One injectable drug resistance

TDR

- Total Drug Resistance
- No Rx

ATYPICAL MYCOBACTERIA

01:16:09

Runyon's Classification

Photo Chromogens	MASK
Scoto Chromogens	SSG
Non Chromogens	MAC, M. xenopi, M. ulcerans
Rapid Growers	M. cheloni, M. fortuitum

Photo Chromogens

- Grow in light

M	Mycobacterium	marinum
A	Mycobacterium	asiaticum
S	Mycobacterium	simiae
K	Mycobacterium	kansasii

Scoto Chromogens

- Grow in darkness

S	Mycobacterium	szulgai
S	Mycobacterium	scrofulaceaum
G	Mycobacterium	gordonae

Non Chromogens

- Neither light nor darkness required
- MAC → Mycobacterium Avium Complex / Mycobacterium Intracellulare aka → BATTEY'S BACILLUS
- M. xenopi
- M. ulcerans

Rapid Growers

- M. cheloni

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PrepLadder

- M. fortuitum
- M. phlei
- M. smegmatis
- M. abscessus

DISEASES

1) Swimming Pool Granuloma/ Fish Tank Granuloma → d/t M. marinum
2) Buruli Ulcer → d/t M. Ulcerans
3) Post Injection Abscess → d/t Rapid Growers
4) Most Atypical bacteria causes → Cutaneous Infections
   Lymphadenopathy

Atypical Mycobacteria which mimics M. TB in Lung involvement → M. Kansassi

MIP: Mycobacterium Indicus Pranii
→ MW Vaccine
→ Vaccine of leprosy
→ Immunomodulatur

In HIV positive (CD 4 < 50)

- MAC
- CMV Retinitis
  - Rx: Ganciclovir

Nine Banded Armadillo

MYCOBACTERIUM LEPRAE

01:26:53

- Also termed as HANSEN'S DISEASE
- Not cultured in Pure culture media
- Doesn't Follow KOCH Postulates → M. leprae, T. pallidum
- Grows in FOOT PAD of ARMADILLO (9 Banded) (best cultured)
- Grows in FOOT PAD of MICE

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PrepLadder

TT	BT	BB	BL	LL
Tubercular Tuberculoid		Border line		Lepromatous leprosy
Paucibacillary			Multibacillary	
– Early involvement of nerve – Granuloma formation in BT – Nerve abscess common in BT – MC in India BT – Good prognosis – BI: 0-1 + – Hypopigmented annular macules				– Late nerve involvement – Legonine facies – Luciophenomena (Periarteritis) – Poor prognosis – BI: 4-6 + – Multiple nodules

*BI: Bacteriological Index

LEPROMIN TEST 01:38:04

- Type IV HSR
- Leopromin Test is a PROGNOSTIC TEST, not a diagnostic test

EARLY LEPROMIN REACTION/ FERNANDEZ REACTION	LATE LEPROMIN REACTION/ MITSUDA REACTION
– read after 3 days – measures Induration Diameter	– reads after 3 wks – measured Nodular Diameter
– Induration › 10mm → +ve	– Nodule › 5 mm → +ve

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LEPRA REACTIONS

TYPE 1 LEPRA REACTION	TYPE 2 LEPRA REACTION
– Down grading & up grading reaction – Type IV hypersensitivity – Mc Feature → edema – mc nerve involved → Ulnar – in BB	– Erythema Nodosum Leprosum – Type III hypersensitivity reactions – immune complex mediated reaction – crops of painful nodules over the skin [d/t TNF] – In BL, LL
– TH1 response ↓ IFNγ & IL-2	– TH2 response ↓ IL-6, 8

PrepLadder

- Can occur spontaneously or after the treatment
- Lepra reaction is a case of emergency

– DOC → Gluco Corticoids
  → Thalidomide can be used in Type 2

Lab Diagnosis

- Slit Skin Smear Examination
  – Taken from:
  – B/L ear lobes
  – Forehead
  – Chin, cheecks
  – Buttocks
  – Nasal mucosal swabs + tissue smear

Cigar bundle leprae

- ZN Staining
  – Add carbol fuschin, do intermittent heating
  – Add 5% H_2So_4
  – Add Methylene Blue
  – Blue background
  – Cigar bundle shape/ Globi like arrangement of leprae seen

- For Prognosis of leprosy
  – Morphological Index → Better
    - Measures % of Solid stained bacilli (Lice bacilli)

Treatment 01:43:02

1) Pauci Bacillary → D + C + R (As per new WHO Update)

- Dapsone
  Clofazimine
  Rifampicin
- For 6 months
- In India: D + R x 6 months

2) Multi Bacillary → D + C + R

- Dapsone

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PrepLadder

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Or click this page to join

Clofazimine

Rifampicin

- For 12 months
- In India: D + C + R x 1 yr

Grading of Smear

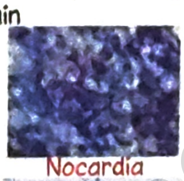
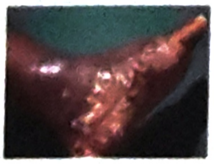
01:51:10

1-10 bacilli/ 100 OIF	→ 1+
1-10 bacilli/ 10 OIF	→ 2+
1-10 bacilli/ 1 OIF	→ 3+
10-100 bacilli/ OIF	→ 4+
10-1000 bacilli/ OIF	→ 5+
>1000 bacilli/ OIF	→ 6+

- Based on this grading, BI is measured

NOCARDIA & ACTINOMYCETES

01:54:40

Nocardia	Actinomycetes
- Strict (Obligate) aerobe - 1% Acid fast - Soil (Exogenous) - MC site: Pulmonary ↓ Pulmonary Nocardiosis - MC extra pulmonary site Brain  Nocardia Antler like branching (Filamentous branching) Actinomycetes	- Strict (Obligate) anaerobe - Mainly non acid fast - Mouth (Endogenous flora) - MC site: Angle of Jaw - Oral-cervical actinomycosis - Triad: Sinus Swelling Sulfur granules ↓ ○ Crush them & see under microscope ○ Sun ray appearance Sulphur Granule - A. israelii ○ Acid fast ○ Leads to PID ○ Occurs in IUCD users
Nocardia & Actinomyces in combination - Actinomycetoma - Multiple sinuses - Mycetoma foot  Mycetoma	
- Culture: Paraffin bait technique	- Spider molar tooth appearance on solid media - Fluffy ball on bottom in liquid media
- DOC: Cotrimoxazole	- Penicillin

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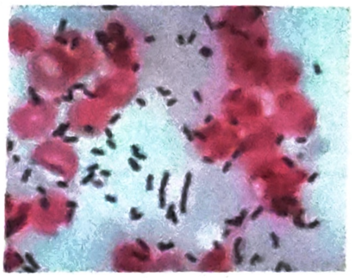
PrepLadder

LISTERIA MONOCYTOGENES

02:04:07

- Gram Positive cocci Bacillus
- Tumbling motility [Peritrichous Flagella]
- Differential motility based on temp. → at 37°C → Non motile
  → 20-25°C → Motile



L. monocytogenes

- Culture
- – Blood Agar
- – Chocolate Agar
- – PALCAM Agar

– Cold ENRICHMENT METHOD [at 4°C]

*In pregnancy → Avoid refrigerated food
↓
May contain listeria
↓
PROM

Reactions:

- ANTON TEST Positive
- CAMP TEST Positive
- CATALASE TEST Positive

ANTON TEST → Putting the Listeria culture into eyes of Rabbit
→ Keratoconjunctivitis

DISEASES

1) Early Neonatal phase → Granulomatosis Infantiseptica
2) Late Neonatal phase → Meningitis
3) In Pregnancy → Chorioamnionitis → PROM
   - ↓ Preterm Births
   - ↓ Abortions
4) In immunocompromised → Sepsis Meningitis
   - DOC → AMPICILLIN
   - It promotes: Actin polymerization for intracellular movement

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PrepLadder

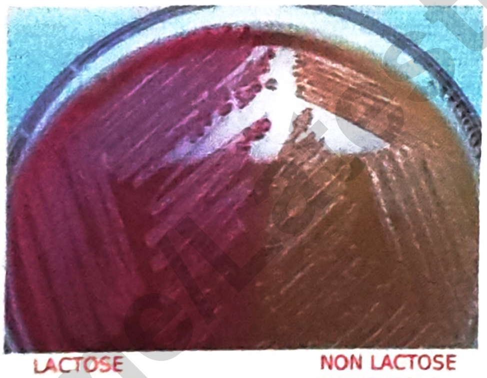
Chapter 5

GRAM NEGATIVE BACILLI -1

ENTEROBATERIACAE FAMILY 00:00:22

Common Features

1) Gram Negative Bacteria
2) Catalase Positive, except: Shigella, Dysentriae- type I
3) Oxidase Negative
4) Nitrate to Nitrite Reduction Test Positive
5) Motile, except: Klebsiella & Shigella, S. gallinarum, S. pullorum
6) Ferment sugar & Produce Acid with gas



LACTOSE FERMENTERS NON LACTOSE FERMENTERS

Lactose Fermenters
E. coli Klebsiella Shigella sonnei

E.coli 00:03:48

	I	M	Vi	C
	Indole	Methyl Red	Voges Proskau er	Citrate
E. coli	Positive	Positive	Negativ e	Negative

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PrepLa

Klebsiella	Negative	Negative	Positive	Positive

*Color change in citrate Test → Green to Blue ["Gram Bacillus"]

DISEASES

1) UTI
2) Diarrhea
3) Neonatal Meningitis
4) Emphysematous Pyelonephritis
5) Fournier's Gangrene
6) Primary Bacteria Peritonitis
7) Scromboid Fish Poisoning
   MC → Proteus > Morganella > E. coli
   Morganella > Proteus > E. coli (overall)
8) Histamine Production increased
9) Pneumonia
10) Liver abscess

UTI

- 'P' Fimbriae possessing E. coli (Mannose Resistant) attach to uroepithelial cells
- Criteria for diagnosis of UTI: > 10^4 CFU/ ml + Symptoms / signs of UTI
- > 10^5 E. coli/ ml of urine → Significant Bacteriuria
- Even 1 bacteria/mL in supra Pubic Aspirate
- > 2 types of colonies on a culture plate → Contamination
  - Then repeat the sample

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PrepLadder

- Mid stream urine sample taken

↓

Mac Conkey/ CLED (Prefrred)

Next Day: › 100 colonies x 10^3 (dilution factor)

↓

› 100

↓

UTI

Diarrhoea Causing E. coli

00:08:48

- EPEC → Entero Pathogenic E. coli
- EIEC → Entero Invasive E. coli
- ETEC → Entero Toxigenic E. coli
- EHEC → Entero Haemorrhagic E. coli
- EAEC → Entero Aggregative E. coli

EPEC

- Causes Pediatric age group Diarrhoea
- Attach to Intestinal villi & Damage them
  - Causes Attaching & Effacing lesions (A/E Lesions)

EIEC

- Resembles Shigellosis
- invasiveness d/t VMA (Virulence Marker Ag)
- Sereny Test Positive

ETEC

- Causes Travelers Diarrhoea
- TOXINS
  - Heat Stable Toxin → acts by ↑ CGMP
  - Heat Labile Toxin → acts by ↑ CAMP
- CFA (Colony Factor Ag)

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PrepL

EHEC

- 0157:H7
  - O → Somatic antigen
  - H → Flagellar antigen
- causes Hemolytic uremic Syndrome & hemorrhagic colitis in children
- TOXINS

  Verotoxin 1 & 2 [Vero Toxigenic E. coli] / SHIGA LIKE TOXIN
- Cuture Media
  - SMAC [Sorbital Mac - Conkey Agar] Does not Ferment sorbital
  - Rainbow agar

EAEC

- Causes Persistent type of Diarrhoea
- Causes Traveler's Diarrhoea too [Few cases]
- Toxin: EAST 1
- Stacked Brick Appearance on Hep 2 cell lines

Neonatal Meningitis

Common causes of Neonatal Meningitis:

- E.coli [with K1 antigen] → MCC
- GBS [Streptococcus agalactiae] → MCC Worldwide

Lab Diagnosis (E. coli)

00:26:21

UTI:

- Wet mount examination
  - Pus cell/ Bacteria
- Leucocyte esterase test
  - L. esterase enzyme in urine pus cells
- Nitrate Reduction Test

  (Griess Nitrite Test)
- Gram staining

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PrepLadder

Confirmatory: culture on CLED/ Mac Conkey agar

↓

Differentiate LF colonies

Sample: Urine/ Stool

Scromboid Fish Poisoning

- a/w Histamine Production
- caused by E. coli › Morganella

  [Note - As per new data → Morganella › Proteus › E. coli]

  - Morganella
    - belongs to family PROTEA
    - Proteus is another member of the family

KLEBSIELLA

00:29:27

- I M Vi C

  \- \- + +

- Green → Blue
- Gram Negative Bacillus
- Capsulated
- Non - motile
- Lactose Fermenter
- Urease Positive [Yellow → Pink]

Urease Test -ve Urease Test +ve

UREASE POSTIVE ORGANISMS	
Staph	→ Staph. aureus
Klub	→ Klebsiella
Main	→ Morganella
Urease	→ Ureaplasma Urealyticum
Programme	→ Proteus
Hai	→ H. Pylori [max. urease producer]
Chalo	→ Cryptococcus neoformans

Klebsiella pneumoniae [Fried Lander's Bacilli]

- CAUSES typical pneumonia (Lobar pneumonia)
- a/w RED Current Jelly Sputum

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Prepl

- a/w Bulging Fissure Sign on CXR

Klebsiella Ozaenae

- a/w ozaena [atrophic rhinitis]
  - Roomy nose
  - Immense foul smell
  - Anosmia
  - Rx by Na HCO_3 douches
    MODIFIED YOUNG'S OPERATION

Klebsiella rhinoscleromatis

- Causes Rhinoscleroma [Woody Nose]
- On Biopsy:
  - MIKULIZ CELLS
  - RUSSEL BODIES

Lab Diagnosis

00:35:26

UTI	Pneumonia	Meningitis
↓	↓	↓
Sample: Urine	Sputum	CSF

Culture: CLED / Mac Conkey Agar / BA

LF Colonies: ↓ Yellow (CLED) ↓ Pinkish (Mac Conkey Agar)

- Very mucoid colonies
- PCR for K. Species
- NDM-1 (New Delhi Metallo-Lactamase)
  Resistance to all types of Antibiotics
- E. coli / Klebsiella → ESBLs enzymes

PROTEUS

00:38:53

- GNB, NLF, Non capsulated
- SWARMS

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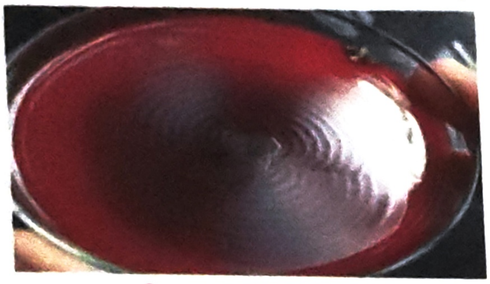
PrepLadder

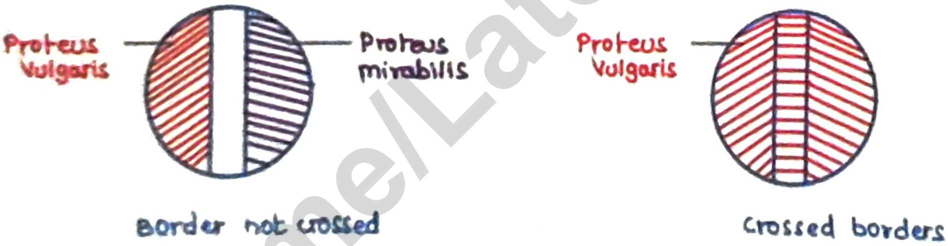
- INHIBIT swarming by
  - ↑% Agar from 2% to 6%
  - instill some chemicals [Boric acid & chloral Hydrate]



Swarming

- Can be cultured on:
  - BA
  - Mac - Conkey Agar → Produce Non Lactose Fermenting [NLF] colonies
  - Phenyl Pyruvic Acid Agar
  - Fishy smell/Seminal smell produced on both media

- Dienes Phenomenon



WEIL – FELIX REACTION

- Proteus Antigen Interactions
  - Ox 19, 2 & K → cross reacts with Rickettsial family members
  - Gives WEIL - FELIX REACTIONS [Heterophile agglutination test]
  - Ox K is strongly positive For scrub typhus
- Proteus a/w chronic UTI:
  Leads to Struvite/Triple Phosphate stones/Staghorn calculus

SALMONELLA

00:46:10

- GNB
- Motile mostly

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Non motile → S. gallinarum
S. pullorum

- Ag's: O, H, Vi
  - O – Somatic
  - H – Flagellar

- KAUFMANN & WHITE SCHEME CLASSIFICATION

	Serogroup	'O' Ag	Vi Ag	H	
				Phase 1	Phase 2
S. paratyphi A	A	1, 2, 12	Absent	a	1, 5
S. paratyphi B	B	1, 4, 5, 12	Absent	b	1, 2
S. typhi murium	B	1, 4, 5, 12	Absent	i	1, 2
S. typhi	D1	9, 12	Present	d	Absent

'O' Antigen	WIDAL	'H' Antigen
– Part of 'LPS' cell wall		– Made of Flagellin
– Heat stable		– Heat labile
– 'O'	WIDAL	– 'H'
– Less immunogenic		– More immunogenic
– 'O' Ab appears early & goes early		– 'H' Ab appears late & goes late
– Granular chalky clump		– Loose fluffy clumps

Variation in 'O' Ag:

- S – R variation

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(Smooth-Rough)

- In Salmonella
  - Lysogenic conversion occurs
  - Bacteriophage mediated
  - Loss or gain of an 'O' Ag

Variation in 'H' AG

- OH-O' variation → Loss of Flagella
- On Phenol Agar
- d/t mutation

Phase Variation

Phase 1	Phase 2
Sero type specific	Non specific

SALMONELLA TYPHI

- Causes TYPHOD FEVER
  - Enteric FEVER caused by
    - S. Typhi
    - S. Paratyphi A, B & C
- Transmitted by Faeco oral contamination
- Infective dose $10^3 - 10^6$ bacilli
- Entry through epithelial cells 'M' cells → Present in intestinal mucosa

Bacteria Mediated Endocytosis

- Process of uptake of bacteria
- Type III secretion system involved
- Following entry bacilli remain in the vacuoles in cytoplasm

ENTERIC FEVER

- Stepladder pattern fever
- ROSE SPOTS Seen

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Prep

- Early abdominal pain, Nausea, Vomiting
- Hepatospleenomegaly, Relative bradycardia → Faget's sign

ROSE SPOTS

COMPLICATIONS

- GI bleed
- Intestinal perforation } 3rd & 4th week of illness
- Longitudinal ulcers seen
- PEA SOUP DIARRHOEA Seen

LAB DIAGNOSIS 01:04:52

B → Blood culture/ Bone marrow culture → 1st week
A → Agglutination Test → 2nd Week
S → Stool culture → 3rd Week
U → Urine Culture → 4th Week

BLOOD CULTURE

- Overall Best
- Blood: culture Fluid → 1:10 [Eg. 10 ml : 100 ml]
  ↓
  Bile broth or Glucose broth can be used
- Add SPS [Sodium Polyanethol Sulfonate]
  – to remove antibiotic effect
- Initial report: 3 days
- Final report: 7 days

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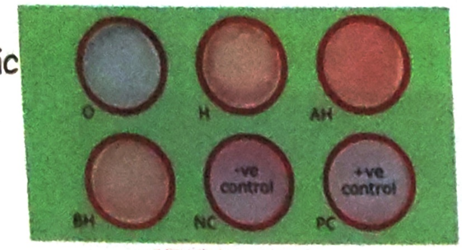
PrepLadder

WIDAL TEST



WIDAL TEST

- Highly sensitive but Poorly specific
  - also positive in BFP – Biological False Positive
    - Infectious mono nucleosis
    - Malaria
    - SLE [auto immune Disease]

- Anti Body titre against O → ›1:100 } Positive
  H → ›1:200

  Flagellar antigen is more immunogenic

- Paired testing should be done, after 2 weeks of initial testing 2nd Test → 4 Fold ↑ in titre should be present → Positive
- IgM Typhi Dot Testing
- Antibody titre values are not fixed, they change according to locality
- Types

1) SLIDE WIDAL TEST
2) TUBE WIDAL TEST
   - Serial dilution with Normal saline

     PROZONE PHENOMENON is avoided
     - Prozone phenomenon → Antibody excess

STOOL & URINE CULTURE

- Enrichment media [SELENITE F BROTH] is used
- CN broth } can also be used
- Tetrathionate broth } can also be used
- Selective media

  DCA [Deoxy cholate Agar]
  XLD [Xylose Lysine Deoxycholate]
  SS [Salmonella Shigella Agar]
  HE [Hektoen enteric agar]
  Welson & Blair Media (Best)

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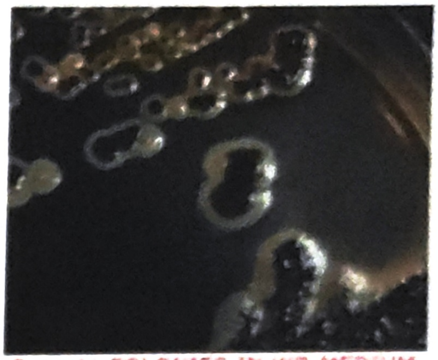
Prep

Above Medias Are also used for Shigella

Except Wilson & Blair Media

- Best media for S. typhi
- Jet Black colonies are Produced



S. typhi COLONIES IN WB-MEDIUM

- Urine culture on → Mac-Conkey agar
  → Cled Agar
- Transport Media → for Salmonella/ Shigella
  → Sach's Buffer Glycerol Saline
- Chronic Carrier
  - >1 yr
  - Harboured mostly in Gall Bladder
  - d/t abundant bile in Gall Bladder

Vi Antigen

- Vi mostly covering up 'O'
- Vi Phage typing can be used For epidemiological studies

NON SALMONELLA TYPHI

- S. choleraesuis → Septicaemia
- S. typhimurium, S. enteridis } Causes Gastro enteritis
- S. typhimurium
  - a/w contaminated Poultry Products
  - can enter the intact egg shell

DOC - 3rd Generation CEPHALOSPORINS

* Reactive artheritis → can be a/w Salmonella, Shigella
* Salmonella a/w Osteomylitis (in Sickle Cell Anemia)

SHIGELLA 01:24:19

- GNB

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PrepLadder

- Catalase +ve, except:
  - S. dysentriae type 1
  - Catalase negative
- Non motile
- Mannitol fermenters

  Except: S. Dysentriae Type 1 →Non mannitol fermenter
- Feco-oral route
- 10-100 bacilli →Infective des
- M' cell play a role here
- VMA:
  - Virulene Marker Ag
  - Invasiveness
- Secretes:
  - Shiga toxin
  - Made by: S. dysentriae type 1 - secretes EXo & Endotoxin
- MCC of Bacillary dysentry:
  - In India: S. flexneri
  - In world: S. sonnei (Late LF)
- Most Shigella species: Serotying
- For S. sonnei: Colicin typing, 26 types

Clinical Manifestations of Shigellosis

- IP: 1-4 days
- Initial Phase: Watery diarrhea

  ↓ Later on

  Dysentry Phase: mucopurulent stools (Bloody)

  Phase of Complications (<5 yrs of age)
  - Tonic megacolon
  - Perforation

  Metabolic Complications:
  - Hypoglycemia
  - Hyponatremia
  - Dehydration

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"EKIRI" SYNDROME

- Toxic Encephalopathy
- Metabolic complications present
- Neurological features
- (Delirium, confusion)

POST INFECTIOUS PHASE:

HLA-B27 → Autoimmune reaction months after infection

↓

Reiter's Syndrome
- Reactive arthritis
- Uretheritis
- Conjunctivitis

Culture Media

- Transport Media: Sach's Buffered Glycerol Saline
- Enriched media → SFB
- Selective media → DCA
  - XLD
  - HE
  - SS

DOC: Ciprofloxacin

NON fermenters - Doesn't Ferment Sugars

1) Pseudomonas
2) Burkholderia mallei
3) Burkholderia Pseudo mallei
4) Acinetobacter baumanii
5) STM

PSEUDOMONAS

01:40:04

- GNB
- catalase +ve
- Oxidase +ve

Oxidase reagent

- Tetra methyl Para phenylene diamine di hydro chloride

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PrepLadder

- Comes as white Disc
- Change of color From white to blue within 10 sec → Positive

- VIRULENCE FACTORS

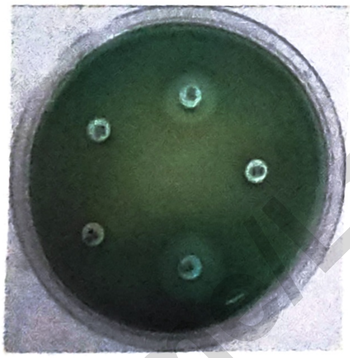
1) Capsular Polysaccharide
2) Alginic Acid- Mucoid strains, Biofilm formation
3) Pigments

a. Pyocyanin	→	Blue
b. Pyorubrin	→	Red
c. Pyomelanin	→	Brown Black
d. Pyoverdin	→	Green

- mc
- mostly found in Pseudomas aeruginosa (mainly Pyocyanin)

These Pigments enhance on KING's Media

Muller Hilton Agar



LAB DIAGNOSIS 01:44:24

- Best selective media → cetrimide Agar

On Mac Conkey- NLF } Fruity odour, Grape Like odour
On NA- Greenish Pigmentation

Pseudomonas Infection

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DISEASES

1) Shanghai Fever
2) Malignant Otitis Externa (Fulminant)
3) Ventilator Associated Pneumonia (MCC)
4) Respiratory infections →in cystic fibrosis
5) Bacteremia
6) Native Valve Endo Carditis
7) Ear infection: Swimmers ear (among children)
8) Eye infection: Corneal ulcer, in contact lens
9) Burn Wound Infections
10) Ecythma Gangrenosum
11) Green Nail Syndrome
   - Inflammation of tissue adjacent to nail
   - Green pus
   - Blue-Green pus
12) Meningitis
13) Bone & Joint infection

Rx: Anti Pseudomonial Drugs

*Vancomycin is not an Anti Pseudomonial
   - Polymixin B & Colistin
   - No Drug resistance reported

BURKHOLDERIA MALLEI

- Causes GLANDER'S DISEASES
   - Animal disease [Horses]
- Non - motile

BURKHOLDERIA PSEUDO MALLEI

- Causes MELIODOSIS
- aka WHITMORE'S BACILLUS
  VEITNAM BOMB
- Both Burkholderia are Bipolar stained/ Safetypin appearance

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PrepLadder

ACINETOBACTER BAUMANII

- GNCB
- Oxidase negative
- Isolated in ICU
- MDR [multi Drug Resistant] isolate
- Oxidase negative
- A. Baumanii
  - OMPP
  - LPS
  - Siderophore
  - Biofilm formation
  - a/w UT, VAP
  - lead to CLABSI (Central Line Associated Blood Stream Infection)
  - a/w Burn Wound Infection

As per WHO → Now-a-days
Acinetobacter infections are increasing

STENOTROPHOMONAS MALTOPHILA

- Saprophytic in plant roots
- Immuno deficient individuals are more prone
- Leads to pneumonia
- Oxidase negative
- Motile
- Oxidatively ferments: Glucose, Maltose & Lactose

VIBRIO

02:03:11

- GNB
- Curved, comma shaped

C –	Catalase	Positive
O –	Oxidase	
I –	Indole	
N –	Nitrate reduction test	
S –	Sucrose lysis	
	String test	

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On the basis of O1

- Classical Vibrio
  - Ogawa
  - Inaba
  - Hikojima
- Eltor Vibrio
  - Ogawa
  - Inaba
  - Hikojima

On the basis of 'O 1' antigen, Vibrio classified into

	Classical	Eltor
Polymyxin B sensitivity	Sensitive	Resistant
Mukherjee Phage IV assay	Sensitive	Resistant
VP [Voges Proskauer]	Negative	Positive
CAMP	Negative	Positive
Chick Erythrocyte Agglutination	Negative	Positive

O2-O139 Vibrosis

→NAG (Non Aglutinuin Vibrio)

O139 →Beugal strain reseculeles Eltor Vib

V. Cholerae

- Virulence factors
  - TCP →Type III finulariae, helps in attachment
    (Toxin Coregulated Pilus)
    (Infective dose: 10^8-10^{10})
  - CT (Cholera Toxin)
  - Zona occludens toxin →disrupts tight junction
  - Siderophore
  - LPS →does not play role in pathogenesis of cholera
  - Used to prepare a killed vaccine

1st 6 Pandemics: by classical strain

7th Pandemics: by Eltor

O139: Bengal outbreak

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PrepLadder

Cholera Toxin

2A + 5B

↓ (2A)

A_1, A_2

↓

ADP ribosylation of GTP

↓

↑ adenylate cyclase activity

↓

↑ CAMP

↓

Intra cellular water in lumen

↓

Massive cholera (Rise watery stool)

↓

Mimics arsenic metal poisoning

↓ (5B)

B-helps in binding to GM_1 ganglioside Receptor

Rx: Rehydration with IV fluids

Lab Diagnosis 02:14:21

- Stool sample
- Hanging drop →Darting motility seen
- String test: Stool + Na deoxycholate →string forms
- Stool sample
  - Put cover slip over it
  - Fish in stream appearance
- Transport media for stool sample
  - VR
  - Carry Blair
- Enrichment media
  - Alkaline peptone water
- Best selective media

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- TCBS
- Greenish yellow
- Monsur's taurocholate, trypticase tellurite agar
- Bile salt agar

RICE WATERY STOOLS

HALOPHILIC VIBRIOS

1) V. Parahemolyticus [7-8% halophilic]
2) V. Vulnificus [8% halophilic]
3) V. Alginolyticus [10% halophilic]

V. PARA HEMOLYTICUS

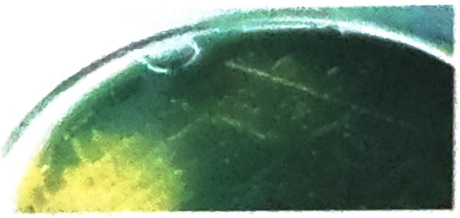
- Capsulated, Bipolar stained
- Causes Sea Food Poisoning (Oyster & crabs)
- a/w Kanagawa Phenomenon on Wagatsuma Agar [enhanced beta hemolysis]
- IP: 8-24hrs

V. vulnificus

- Coral cut wound infection

*TCBS → Non sucrose fermenter

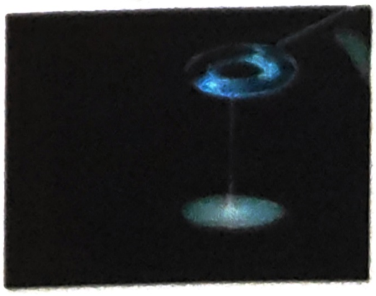
V.Parahemolyticus



TCBS

- STRING TEST

- Sodium deoxycholate [Bile salt] is added to stool sample
- Try to Lift with loop → String Is formed



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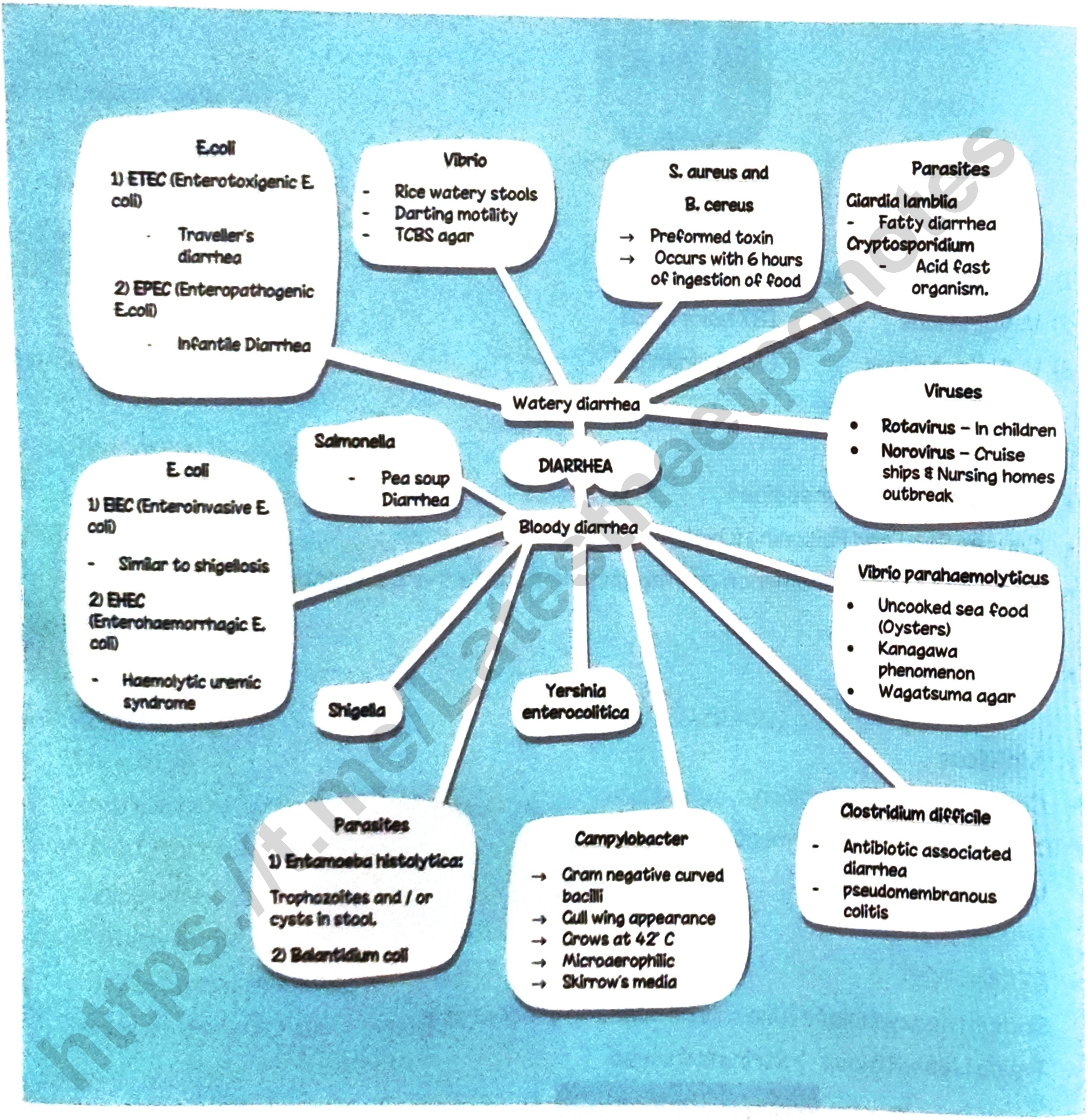
PrepLadder



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PrepLadder

Chapter 6

GRAM NEGATIVE BACILLI - 2

HAEMOPHILUS DUCREYI

00:00:14

- Causes chancroid (soft sore)
  - Painful, soft genital ulcer

*Painless, hard ulcer → Syphilis

Lab Diagnosis:

Tissue biopsy
+
Stain
↓
School of fish appearance
Or
Tram track appearance

- Culture media: on chocolate agar with isovitale-X
- DOC: Azithromycin

HAEMOPHILUS INFLUENZAE

- Termed as Pffeir's Bacillus

X & V factors

X → Hemin

V → NAD

Hemin & NAD: Both present in Chocolate Agar

Modifications of Chocolate Agar:

- Levinthal & Filde's Agar

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PrepLadder

- Capsulated
- Catalase +ve
- Oxidase +ve
- Pleomorphism

Virulence Factors

- Capsular polysaccharide
- Ig A_1 protease
- Endotoxins
- OMP's
- Pili

CLINICAL MANIFESTATIONS

Capsulated	Non Capsulated
• Causes invasive disease ↓ – Pyogenic meningitis – Subdural effusion ↓ Mortality rate high – Epiglotitis – Pneumonia in infants	• Non invasive disease ↓ – Otitis media – LRTI – Pneumonia in adults – Puerperal sepsis

- On the basis of Capsular Polysaccharides, 6 Serogroups are there: a, b, c, d, e, f

- Type 'b' PRP (Polyribitol Phosphate)
  ↓
  Hib Vaccine

SATELLITISM

Satellitism

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PrepLadder

- Shown by H. influenzae

Lab Diagnosis:

d/t H. influenzae → C.S.F should not be refrigerated
→ Cold labile

- Gm staining : Gm negative coccobacilli
↓
Shows pleomorphism
- Capsule detection: Quellung, reaction
- Ag detection: mostly available for Type 'b' capsular Ag
- Culture: BA, CA, Levinthal & Filde's Agar
- Haemophilus Selective Media

H. Influenzae ↓ Does not ferment sucrose	H. parainfluenzae ↓ Ferments sucrose

H. AEGYPTICUS

00:17:37

- Also termed as: Koch Week's Bacillus
- Causes

1) PINK EYES

2) BRAZILIAN PURPURIC FEVER

NORMAL MOUTH COMMENSALS
H → H. parainfluenzae, H. aprophilus, H. para aprophilus A → Aggregatibacter [Actinobacillus myceticomitans] C → Cardiobacterium hominis E → Eikenella coordens [a/w human bites] K → Kingella Kingae They can cause culture negative Endocarditis in immuno compromised

BORDETELLA PERTUSIS

00:20:32

- Causes pertusis/ whooping cough/ 100 day cough/ Barking dog cough

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PrepLadder

Virulence Factors

i) Tracheal cytotoxin
- Part of cell wall
- Damage to cilia of respiratory epithelium

ii) Pertusis toxin

iii) Adenylate toxin

iv) Dermonecrotic toxin → Respiratory mucosal damage

v) Endotoxin (LPS)

vi) Adhesins
- Filamentous haemagglutinin
- Pertactin (OMP)
- Fimbriae
- BrKA Protein → gives B. pertusis resistance to killing & it's adhesion

Acellular Pertussis Vaccine

- Used Now [equal In Efficacy With Cellular Vaccine]
- Made Up Of:
  - Pertussis Toxoid
  - Filamentous Haemagglutinin
  - Pertactin
  - Agglutinogens 1, 2 & 3
  - Fimbrial part

Clinical Manifestations

- IP 1-2 wks

STAGES

- Cattarhal Phase
- Paroxysmal phase
- Convalescent phase

Cattarhal Phase: 1-2 hrs

- Non specific features: Common cold, cough (mild)

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PrepLadder

- Culture positive

Paroxysmal phase:

"Whoops" present

- Whooping cough
- Post tussive vomiting
- Culture negative

Convalescent / Recovery Phase

- Abs may be present in the serum

Lab Diagnosis:

- Nasopharyngeal swab (6 swabs)
  - Gm stainig
  - Culture

Culture Media:

- Bordet Gengou Media
  - Bisected pearl appearance
  - Mercury drop appearance or Thumb print appearance on culture smear
- Regan - Lowe media

DPT Vaccine

- Pertusis Acellular component
- Adjuvant
- Increase immunogenicity of whole vaccine

DOC: Macrolides

BRUCELLA

00:36:19

- Zoonotic Disease

MOT:

- by eating & drinking contaminated food & milk products

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PrepLadder

- contact with animals (aerosols)

CAUSES

B. abortus → infects cattles
B. melitensis → infects sheep, Goat, camel
B. suis → infects Pigs
B. canis → Infects Dogs

B. MELITENSIS

- Causes Acute Brucellosis/ Malta/ Meditteranean/ Undulent Fever
  - Typhoid Like illness
  - Fever, Rash, Hepatospleenomegaly, Arthritis
- Undulating Nature of fever: Typical remittent course
  ↓
  Recovery phase between 2 febrile episodes
  - Vertebral osteomyelitis involving lumbar region
  - Septic arthritis

Lab Diagnosis:

Specimens: Blood, Bone Marrow Culture

- Blood Culture: Castaneda method of isolation
  - Ratio of blood & culture fluid → 1:10
  - Glucose Broth is taken as culture Fluid
  - Add ERYTHRITOL → Stimulate Brucella growth
  - Carried by CASTANEDA METHOD
    - o Biphasic media [Solid & liquid culture media]
    - o use - minimizes contamination in Sub cultures
  - Gm staining: GNCB

- Catalase
- Oxidase
- Urease

} Positive

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- Tblisi Phage Typing
- Serological Test:

i) SAT →Standard Agglutination Test →Detects IgM Ab

ii) 2 ME (Mercapto Ethanol) Test →Done for IgM Ab

iii) PCR for Brucella species

TEST FOR BRUCELLA CONTAMINATION

* Brucella contaminates milk

i) Rose Begnal card test

ii) Milk ring test

iii) Whey Agglutination Test

DOC: Rifampicin + Doxycycline (WHO)

Streptomycin + Doxycycline (Harrison)

YERSINIA PESTIS 00:47:59

- Leads to Plague (Zoonotic infection)

Culture Media

- Ghee broth
- Stalactite type of growth

Virulence Factors

i) F1 Ag → Plasmid borne

→ Heat labile capsular Ag (inhibit macrophage phagocytosis)

ii) Phospholipase

iii) PH 6 Ag → Fibrilar protein

→ Binding to lipoprotein

iv) LPS → Endotoxic activity

v) Siderophore

- TYPES

1) B → Bubonic → IP → 2-7 Days

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PrepLadder

2) S → Septicaemic → IP → 2-7 Days
3) P → Pneumonic → IP → 1-3 Days [most Dangerous]

BUBONIC PLAGUE

- MC type
- MOT: Bite (Rat Flea)

  ↓

  Lymphatics

  ↓

  Multiplies
- IP: 2-7 days
- Buboes: Regional LN

  Tense & Tender
- Bubonic Plague can't spread from Person to Person

PNEUMONIC PLAGUE

- IP: 1-3 days
- MOT: Inhalation of bacilli in droplets expelled from another person or an animal
- Rare: < 1%
- Agent of Bioterrorism

Clinical Features

Fever, headache & pneumonia like features

SEPTICAEMIC PLAGUE

- LPS: Endotoxic shock
- IP: 2-7 days
- Massive involvement of blood vessels → Gangrene → Black death

Lab Diagnosis

Bubonic	→ Buboes smear (Pus or Fluid)
Pneumonic	→ sputum & blood
Septicaemic	→ Blood & spleenic aspirate

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PrepLadder

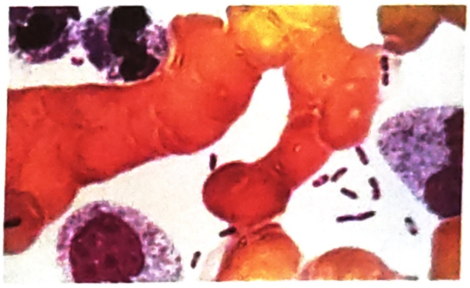
Transport Media: Carry Blair

Direct Microscopy

- Presence of pus cells & GNCB
- Wayson stain or Methylene blue stain
  - Bipolar stained appearance

    or

    Safety pin appearance



Bipolar Staining

Culture:

- Ghee broth → stalactite growth
- Pleomorphism is also shown
- F1 Ag detection
- PCR for Y. Pestis

DOC: Streptomycin

Prophylaxis: Doxycyclin

- "Cheopis Index" → for plague

Vaccine

- Haffkine vaccine
  - Sokhey's modification (killed vaccine)
  - Live attenuated is also there but not routinely given

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YERSINIOSIS

(Zoonotic Infection)

Y. enterocolitica
↓
Virulence Factors
i) Invasive protein (Invasin Protein)
ii) Yersinia adhesin A (Yad A)
iii) PH6 Ag, MyF Ag

Y. PseudoTB
↓
Y. PseudoTB + enterocolitica
↓
Super Antigen

Clinical Features

i) Self limiting Gastroenteritis

ii) Intestinal complications:

- Terminal ileitis (mimics appendicitis)
  (Y. enterocolitica > Y. pseudoTB)

iii) Septicaemia

iv) Post infective phenomenon

- Reactive arthritis (HLA B 27 positive)
- Erythema nodosum (HLA B 27 negative)
- Grave's disease → Y. enterocolitica mimics TSH binding site

- Specific Culture Media: CIN yersinia agar
  (Cefsulodin Irgasan Novabiocin)
- Cold enrichment at 40°C
- They show, Differential motility
  Motile at 22°C
  Non motile at 37°C
- PCR for various yersinia species

LEGIONELLA PNEUMOPHILA

01:14:05

- Most common serotype
- MOT
  1) Aerosol route from centralized AC plants
  2) Drinking contaminated water with Legionella

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PrepLadder

- DISEASES

1) PONTAIC FEVER

2) Leggionare's Pneumonia

– Complicates with
Encephalitis & Diarrhoea →LEGIONNARE'S DISEASES
- a/w mortality

- CULTURE MEDIA → Buffered Charcoal Yeast Extract [BCYE]
- PCR for Legionella Species
- In Urine: Legionella Ag can be detected
- Fluorescent microscopy
- Legionella micadedi is Acid Fast in nature

DOC: Macrolides

SPIROCHETES

01:19:55

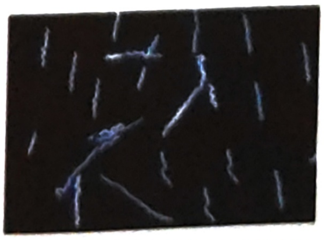
- Spiral in shape
- It includes: Treponema
  Borrelia
  Leprospira

Composition:

- Outer membrane
- Periplasmic space →contains flagella →Endo flagella
- Peptidoglycan layer
- Inner cytoplasmic membrane

Microscopy

- DARK FIELD MICROSCOPY is done
  – Background is dark
  – Reflected light is used



Dark field Microscopy

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PrepLadder

- FONTANA & LEVADITI STAINING done
  - Silver impregnation method
  - makes them thick

	Treponema	Borrelia	Leptospira
Spirals	6-12	3-10	Numerous Tightly coiled Hooked ends
Endoflagella	3-4	7-11	1

TREPONEMA PALLIDUM

- Causes SYPHILIS
- IP → 9-90 Days

STAGES

1) Primary → Hard, painless chancres, painless LAP

2) Secondary → Rashes over the palms & soles are present
→ Condyloma lata lesions are seen
- Most infectious lesion of syphilis

3) Latent → for years

4) Teritary → GUMMAS [skin lesions] seen
→ Complications:
- Neuro syphilis
  Presents with:
  - GPI [General Paralysis of Insane]
  - Tabes dorsalis
- Cardio vascular syphilis
  Presents with Ascending Aneurysm of Aorta

Lab Diagnosis:

- Microscopy : Dark ground
- Direct IF staining
- Silver impregnated methods: L & F
- Non culture can be cultured on Smith Noguchi media

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PrepLa

Serology:

1) NON TREPONEMAL TESTS

a. VDRL [Veneral Disease Research Lab]

b. RPR [Rapid Plasma Reagent]

VDRL	RPR
• Slide Flocculation Test • Needs a microscope • Fluid is to be prepared & to be used in 24 hrs • Preheating of serum • Culture on: Blood, Serum, CSF • Done with CSF [Neuro syphilis] Best test to assess prognosis • IOC for Neurosyphilis: VDRL - CSF • Cheaper	• Better Test – Card test – no Fluid is prepared • On Blood, Serum • in CSF sample, RPR Fails • Costly

2) TREPONEMAL TESTS (More Specific Test)

a. FTA – ABS [Fluorescent Treponemal Antibody Assay]

b. TPI [Treponema pallidum Immobolization Assay]

c. TPHA [Treponema pallidum Haemagglutination Assay]

d. TPPA [Treponema pallidum Particulate agglutination Assay]

MOST SENSITIVE → FTA - ABS
MOST SPECIFIC → TPI
- but routinely not done
- d/t involvement of live Treponemes

→ TPPA
- 2nd most specific Test

MC Done in LABS → TPHA

What is the IOC for Neurosyphilis?

- FTA-ABS CSF
- VDRL CSF (Better Option)

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PrepLadder

IOC for Congenital Syphilis

- IgM FTA ABS is done

Treatment

- DOC for Primary, Secondary & Latent stages of Syphilis → Benzathine Penicillin
- DOC for Neurosyphilis → Penicillin > Procaine Penicillin

Non Treponemal Pallidum T. Pertunae → causes YAWS T. endemicum → causes BEJEL [endemic syphilis] T. carateum → causes PINTA } Not STDs

	YAWS	ENDEMIC SYPHILIS	PINTA
MOT	Skin to Skin	Kissing Sharing utensils	Skin to Skin
	Early childhood	Early childhood	Late childhood
	Ulcerative papilloma	Rarely seen	Non-ulcerating papule
Sites	Extremities	Oral	Extremities Face
Relapses	Common	-	None

BORRELIA

- Epidemic Louse Born Relapsing Fever (Crushing/ rubbing)
  → caused by B. recurrentis
- Endemic Tick Born Relapsing Fever (Tick bite)
  → caused by B. duttoni & B. hermesii

Relapsing Fever:

- Relapses are d/t Antigenic Variations
- B. reccurenting undergo Antigenic variation

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PrepLadder

- Last for 3-5 days

Diagnosis: PCR for Borrelia species

B. BURGDORFERI

- Transmitted by Ticks
- Causes LYME'S Disease
  - Characteristic lesion → Erythema Chronicum Migrans aka Annular Bulls Eye Pattern Rash
- Culture Media → Barbour Stannour Kelly Media

Lyme's Disease

Stage 1

- Early localized infection
- IP: 3-30 days
- Annular maculo papular lesion
  - Develops at tick bite site
- Erythema migrans
  - Over thigh, groin

Stage 2

- Early dissemination
- Blood Borne
- Secondary annular skin lesions
- Arthralgia
- Profound fatigue
- Neurological features
  - Bannwarth's syndrome
    - Meningitis
    - Typical Lymphocytic meningoradiculitis
- Cardiac Involvement

Stage 3

- Late persistent infection
- Lyme's arthritis

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PrepLadder

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- Large Joints involved
- Acrodermatitis Chronic Atrophicans
  (Late skin manifestation)
- Post Lyme Syndrome: aka Chronic Fatigue Syndrome

Lab Diagnosis

1) CULTURE
   - Blood & CSF
   - BSK media or Kelley's media
2) PCR for Borrelia
3) Dark field microscopy

Treatment: DOC → Doxycycline (for all stages)

Except in CNS/ CVS involvement: DOC → Ceftriaxone

B. VINCENTI & FUSIFORMIS

- Causes Vincent Angina / Trench Mouth
- Inflamed pharyngeal mucosa covered by greyish membrane
- DOC: Penicillin, Metronidazole

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Chapter 7

Gram Negative Bacilli - 3

LEPTOSPIRA

00:00:15

- Causes WEIL's Disease [IcteroHaemorrhagic Fever]
- L. interrogens (Most pathogenic for humans)
- L. biflexa

LEPTOSPIRA INTERROGENS

00:03:00

MOT

- Zoonotic Transmission:

  R → Rat urine
  R → Rice [Paddy Field workers
  R → Rainy water

  } a/w LEPTOSPIROSIS

- No Human → Human transmission

Clinical Stages

- Mild illness

	First Stage	Second Stage
	– Septicaemic phase	– Immune phase
	– 3-10 days	-
Features	– Fever, Myalgia, Conjunctival suffusion, Abdominal pain, Vomiting	– Meningitis, Uveitis, Chorioretinitis, Rash, Fever, Peripheral neuropathy

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PrepLadder

Isolation	– Blood & CSF	– From urine
Serum IgM	– Absent	– Present
Antibiotics	– Doxycycline	– Refractory to Rx

- Severe illness → Weil's Disease 00:07:45

	First Stage	Second Stage
	– Septicaemic phase	– Immune phase
	– 3-10 days	– 10-30 days
Features	– High grade fever – Liver → Jaundice & increased Liver enzymes – Hemorrhages o Pulmonary o Skin o Conjunctival – Kidney → Raised creatinine level	– All features of septicaemic phase can be present
Isolation	– Blood & CSF – No IgM – Sensitive to antibiotics	– Urine – IgM present – Refractory to antibiotics

Lab Diagnosis:

- Specimens: CSF, Blood, urine
- Microscopy: Dark ground Microscopy
- Isolation from culture media's: EMJH, Fletcher, Korthof
- Animal inoculation → Not done now
- Serological test: MAT → Microscopic Agglutination Test

PCR for leptospira species (IOC)

DOC: Doxycycline
Penicillin for severe cases

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PrepL

Dinger's ring: Leptospira produces a ring of organism, just under surface of medium

Faine's criteria (WHO approved guidelines)

- For diagnosis of leptospirosis
- Epidemiological + Lab findings
- Non specific findings:
  - Altered renal functions
  - Altered liver functions
  - Urinary sediments

*National Referalcentre for Leptospira located at → Port Blair

RICKETTSIAE

00:22:19

- Obligate intracellular organisms
- Arthropod borne Transmission
- Non culturable
- Fever, Rash, LAP

	CAUSED BY	ARTHOPODS INVOLVED
Epidemic Typhus	R. prowazekii	Louse
Endemic Typhus	R. typhi	Flea
RMSF	R. rickettsi	Tick
Indian Tick Typhus	R. conori	Tick
R. pox	R. akari	Mite
Scrub Typhus	Orientia tsutsugamushi	Mite

- Presents with fever, rash & lymphadenopathy

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PrepLadder

- DOC → DOXYCYCLIN
- Target Site: Endothelial Cells → R. akari, O. tsutsugamushi
  → Can attack monocytes also

Mite

Tick

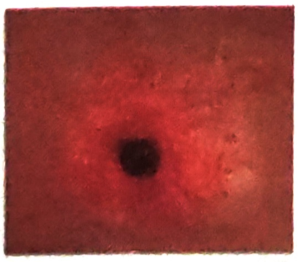
Flea

TUNICA REACTION [NEIL MOOSER'S REACTION] 00:29:25

- Epidemic (-ve) Vs Endemic (+ve) Typhus
  - Scrotal necrosis & inflammation of guinea Pig: Endemic typhus
  - No such Reaction & inflammation in guinea pig : Epidemic typhus
- OX19 & OX2 +ve for both
- These are non motile strains of proteus, cross reacts with Rickettsial Ab's →Weil-Felix reaction
- OXK is strongly positive in scrub typhus

Scrub Typhus 00:32:33

- By O. tsutsugamushi
- Transmitted by Chiggerosis larvae



Scrub typhus

Clinical Manifestations

Triad: Black Escher
Regional LAP
Maculopapular rash

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Non Specific Features: Fever, Headache, Myalgia

Hilly region: Zoonotic Tetrad

i) Trombiculid mite

ii) Mice, rats

iii) Scrub vegetations

iv) Wet season

- Strongly +ve for OXK

  DOC: Doxycycline

  PCR for Scrub typhus

Q FEVER

00:38:16

- Caused by Coxiella burnetti
- Contaminates milk
  - Killed by Flash method of Pasteurisation
- MOT → Aerosol transmission [no arthopod transmission]
- No Rash occurs
- Non cultured
- a/w culture negative endocarditis

Pathogenesis

- Escapes intracellular killing by inhibiting final step of phagosome maturation
- Resist acidification of Phagolysosomes

Clinical Manifestations

- Acute 'Q' fever
- IP: 3-30 days
- Hepatitis, pneumonia, fever
- CNS & CVS involvement may be seen
- Post 'Q' Fever Fatigue Syndrome
- Latency → can be for 2-3 days
- Chronic 'Q' Fever
  - a/w Endocarditis

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PrepLadder

- fever mostly absent

Phase Variation

Phase 1 →Fresh isolates (LPS surface)

On repeated passage in Yolk sac

↓ Surface Protein Enclosed

Phase 2

COXIELLA BURNETTI

- GNCB
- Pleomorphic
- Isolation: in BSL – 3 lab
- Ab detection:
  - Indirect IF testing
  - IgM: appears in 7-10 days
  - IgG: appears in 14-20 days of infection

In Chronic Infection:

IgG to Phase 1 →Shows elevated levels

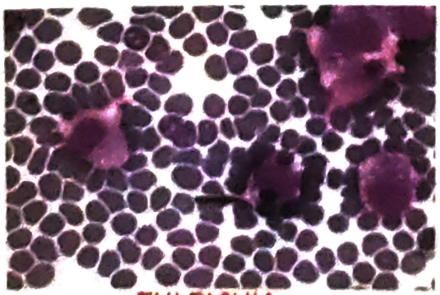
→› 1:6400

- CFT
- PCR for C. burnetti
- DOC: Doxycycline

EHRLICHIA

00:48:36

E. chaffeensis	E. ewingii	Anaplasma Phagocytophilum	Neorickettsia Sennetis



EHLRICHIA

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PrepL

E. CHAFFEENSIS

- Causes HME (Human Monocytic Ehrilichiosis)
- Leukopenia
  Thrombocytopenia
  Elevated Liver Enzymes
- Transmitted by Tick

E. EWINGIL

- Causes HGE (Human Granulocytic Ehrilichiosis)
- Features same as E. chaffenesis, but less severe
- Transmitted by tick

ANAPLASMA PHAGOCYTOPHILUM

- Cuases HGA (Human Granulocytic Anaplasmosis)
- Leucopenia
  Thrombocytopenia
- Transmitted by tick

NEORICKETSIA SENNETIS

- Causes HLE (Human Lymphocytic Ehrlichiosis)
- Mononucleosis Like Syndrome
- Transmitted by Fish Ingestion (infected flukes)

BARTONELLA

00:54:50

- Can be cultured on normal culture media

- **B. henselae**
  - Caused CAT Scratch Disease
    →Regional LAP
    →Painless Erythmatouspapule
  - Caused Bacillary Angiomatosis [common in HIV +]→leads to Neovascularisation
  - Hepatosplenic lesions

- Bacillary peliosis

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PrepLadder

- Angioproliferative disorder involving liver
- Bacteriaemia& Endocarditis →Rare association

- B. bacilliformis→causes OROYA FEVER

EhrlichiaChaffenensis →causes Human monocytic ehrlichiosis

EnrlichiaPhagocytophila→causes Human granulocytic ehrlichiosis

- B. Quintana
  - Causes BACILLARY ANGIOMATOSIS [Fever cases]
  - Causes TRENCH FEVER/ 5 day fever
  - By louse feces
  - Seen in WW-I in 1919

- B. bacilliformis
  - Transmitted by sandfly
  - Also termed as Lutzomyia
  - Leads to oroya fever or Carrion's disease
    - Initial bacteremia
    - Systemic illness can happen

- Lately →verruga peruara
  - Cutaneous vascular lesion

Rx for:

Bacillary angiomatosis: Erythromycin Doxycyclin

Cat scratch disease: Erythromycin

Trench fever: Gentamycin

CHLAMYDIA (ATP PARASITE)

01:03:00

- Obligate intracellular organism

Exists in:

- Elementary body - Extracellular from - Infectious	- Reticulate body - Intracellular form - Replicating

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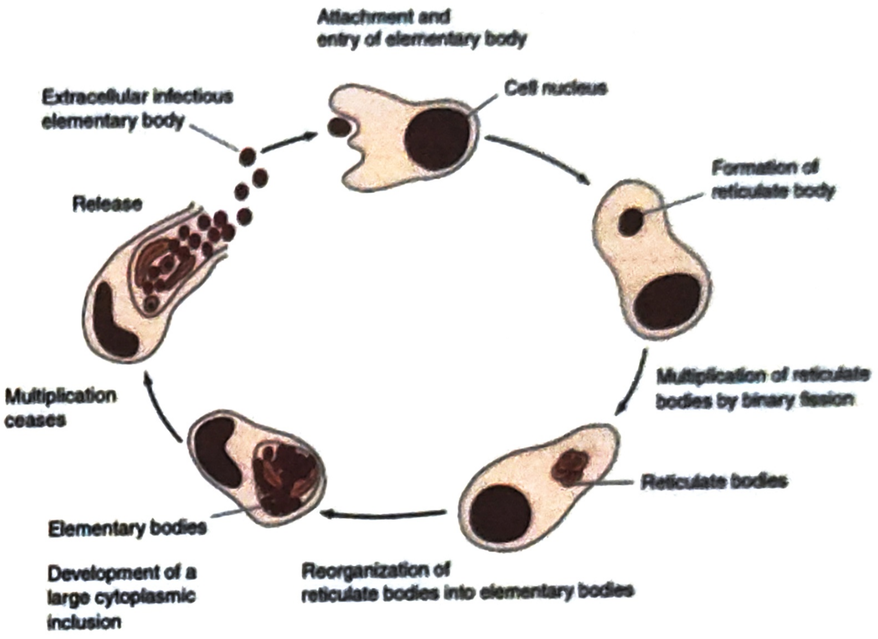
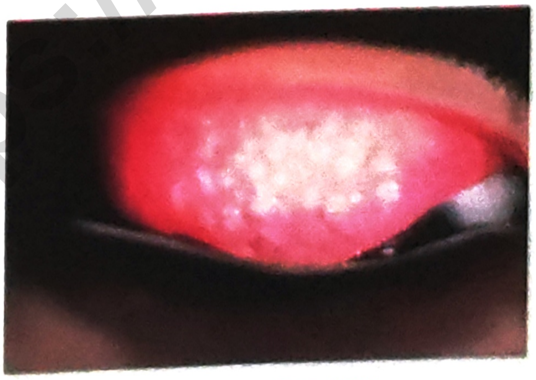


Elementary And Reticulate Bodies

- Termed as: Basophilic viruses
- Pass through bacterial filter
- Non cultured
- Obligate intracellular
- ATP parasite
- Responds to the Antibiotics
- Multiply by binary fission
- Contains 70s ribosome

C. TRACHOMATIS



TRACHOMATIS

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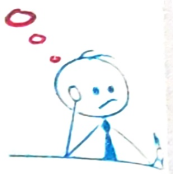
PrepLadder



- Serogroups A,B,Ba,C →causes TRACHOMA
  Mx Strategy
  - S → Surgery
  - A → Antibiotics
  - F → Face clean
  - E → Environmental Clean

- SerogroupsD-K →causes INCLUSION CONJUNCTIVITIS
  INFANTILE PNEUMONIA
  GENITAL CHLAMYDIASIS

- SerotypesL_1,L_2,L_3 → causes LYMPHO GRANULOMA VENERUM [L2>L1]
- Trick agents: A, B, Ba, C, L_2> L_1&D-K

C. PSITTACI

- causes BIRD PNEUMONIA
  - mainly to parrots
    - helps in aerosol transmission

Inclusion bodies: LCL (Levinthal ColeLilee Bodies)

Rx:

- Tetracycline
- Replaced by Doxycycline

C. PNEUMONIAE

- causes ADULT PNEUMONIA
  - TWAR strain
- a/w Atherosclerosis

INCLUSION BODIES	
Trachoma	→ HP Bodies [HALBERDT PROWAZEK]
Molluscum contagiosum	→ HP Bodies [HENDERSON PETERSON]
Psittacosis	→ LEVINTHAL COLE LILEE BODIES
LGV	→ Miyagaviacorpusdes

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PrepL

LGV

- Painless ulcer
- Painful lymph nodes → Para rectal LN involved
- FRIE'S SKIN TEST is done
- Complication → ESTHIOMENE → Rectal & vulval strictures
- $L_2 > L_1$

  A → Asymptomatic

  B → Buboes

  C → C. trachomatis

  D → DOC: Doxycyclin

  E → Esthiomene (Rectal & Vulval Strictures)

  F → Frie's test

  G → Groove sign

- CULTURE
  - on Mcoy cell line [continous cell line]
  - on Yolk sac of eggs [Hen's]

- IOC → NAAT [Nucleic Acid Amplification Test] → ≅PCR

GENITAL CHLAMYDIASIS

- Done by D & K serotype
- Causes NGV → 30-50% MCC
- IP: 7-10 days
- Mucopurulent discharge
- Dysuria & urethral irritation

Reactive arthritis

- a/w Reiter's syndrome
- occurs in 1-2% NGV cases
- M > F
- HLA-B27 strong association

Inclusion Conjunctivitis

- Looks like ophthalmia neonatorum

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PrepLadder

- C. trachomatis > Gonococci
- IP: 6-21 days
- Mucopurulent discharge

Adult Inclusion Conjunctivitis

- Follicular conjunctivitis
- Swimming pool conjunctivitis

Infant Pneumonia

- 3 wks to 3 months of birth
- Infection develops in conjunctiva → Pharynx
- Infection through Eustachian tube → Otitis media

Gm staining

- Gm –ve (Poorly stained)
- Sterile Pyuria (No organism)

IOC: NAAT/PCR for Chlamydia species

Rx:

- Adult conjunctivitis: DOC Azithromycin
- Uncomplicated genital infection: Azithromycin
- Complicated genital infection: Doxycycline or Erythromycin
- Ophthalmia Neonatorum & Infant Pneumonia:
  - Macrolide
  - Erythromycin
  - Topical ointment

* Mcoy cell lines: preferred cell lines on which Chlamydia prefers to grow

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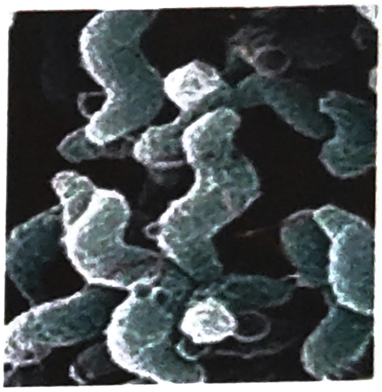
CAMPYLOBACTER

01:26:26

- GNB
- Gull wing shape
- Micro aerophile
- Motile → Darting motility

C. jejuni



- a/w causing the contamination of Poultry products
- Leads to Diarrheal episodes
- In Children
  - Diarrheal episodes

    ↓ f/b

    GBS

Lab Diagnosis:

Stool sample

↓

Wetmount

- Fecal leucocytes present
- Inflammatory form of diarrhea

Gm staining:

- GNB
- Gullwing shape
- Leucocytes

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PrepLadder

Cultured on:

- Campy BAP
- Butzler
- Skirrow

Incubation temp →42°C

HELICOBACTER

01:31:38

- GNB, curved
- Normal commensal of stomach

H. pylori

- 50% of world human population
- Motility →Motile
- Acid resistance d/to:
  - urease enzyme

    Urea $\xrightarrow{Urease}$ NH3 →Neutralizes the gastric acid
  - Amidase & Arginase

    ↓

    NH_3
- Adhesins (mucus layer)
- VaCA gene →Vacoulating cytotoxin
- CagA gene →Pathogenicity islands codes for type II secretion system

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Prep

- These genes make H. pylori more pathogenic

Disease by H. pylori

- Antral gastritis
- Peptic ulcer disease
  - 80% Duodenal ulcer
  - 60% Gastric ulcer
- Chronic atrophic gastritis
- Autoimmune gastritis
- Pernicious anemia
- Adeno carcinoma of stomach
- Non Hodgkin's gastric lymphoma

Protective role of H. pylori

A → Adenocarcinoma of Esophagus
B → Barret's esophagus
G → GERD
Analysis → Asthma

Lab Diagnosis:

Non invasive test

- Urea breath test
  - Sensitive

Invasive procedures

- Endoscopic biopsy
  - Site: Antrum
  - Biopsy tissue + Warthin StarrySilver Stain
  - Gastric biopsy tissue
    - Rapid urease test
    - Highly sensitive

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Culture Media: Skirrow media

Biochemical test:

- Catalase
- Oxidase
- Urease

} +ve

- PCR for H. pylori

Rx Regimes:

- OCA Omiperazole/ Clarithomycin/ Amoxicillin
- OBM Omiperazole/ Bismuth salicylate/ Metronidazole
- OBTM Omiperazole/ Bismuth salicylate/ Tetracyclin/Metronidazole

} x weeks

- Helicobacter/ Pylokits are available

Rat Bite Fever 01:44:48

- High grad fever
- Rashes
- Painful polyarthritis

Done by:

Streptobacillus Moniliformis	Spirillum minus
↓ Normal flora in throat of rats ↓ Leads to Haverhill Fever	↓ SodokuFever
IP: 7-10 days	1-3 weeks
DOC: Penicillin	Penicillin

FRANCISELLA TULARENSIS

- Causes Tularemia
- Plague like disease of rodents
- Zoonotic infections

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Prep

- MOT: contact with wild/ domestic animals

Clinical manifestations

1. Ulceroglandular Tularemia (MC)
2. Pulmonary Tularemia
3. Oropharyngeal Tularemia
   - Also used as Agent of Bioterrorism → Category A
   - Highly infectious

Lab Diagnosis

- Difficult to Culture
- PCR for F. tularensis
- GNCB, Catalse +ve, Oxidase −ve
- Bipolar stained appearance

DOC: Gentamicin

MYCOPLASMA

01:51:38

- Smallest bacteria
- No cell wall
- Cell membrane is present → made of sterol
- Can pass through bacterial filters

Culture media:

PPLD Agar → Fried Egg colonies
Stained by Diene's METHOD
[DIENE'S PHENOMENON seen in PROTEUS]

Fried Egg colonies

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PrepLadder

M. PNEUMONIAE/EATON'S AGENT

- MC bacterial cause of Atypical pneumonia
- aka WALKING PNEUMONIA

Lab Diagonosis:

1) CAT [cold Agglutination Test] → type of Heterophile Agglutination test
2) Streptococcus MG Agglutination Test → Type Of Heterophile Agglutination Test
3) PCR for M. pneumoniae

Culture Media

- PPLO broth
- SPH medium
- Hayflick modified medium
- Diene's method of staining
- CAT
- Diagnosed by PCR

SPUTUM:

- Throat swabs
- NP swabs

Antigenic detection

- Direct IF testing
- PCR for mycoplasma species

NON GONOCOCCAL URETHRITIS CAUSES
1) Chlamydia trachomatis 2) Ureaplasmaurealyticum 3) Mycoplasma genitalium 4) Mycoplasma hominis

Extra Pulmonary Manifestation of M. pneumonia

- Aka Walking Pneumonia
- Neurological

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Or click this page to join

- Meningoencephalitis
- GBS
- Aseptic Meningitis

- Dermatological
  - Skin rashes
  - Erythema multiforme (major)
- Cardiac
  - Myocarditis
  - Pericarditis
- Rheumatology
  - Reactive arthritis
- Hematological
  - Anemia & Hypercoagulopathy

M. genitalium & M. hominis

- a/w Non Gonococcal Urethritis
- MCC → Uroplasma Urealyticum
- MCC → C. trachomatis
- Also a/w Reiter's syndrome

DOC: Macrolides

CAT → Mycoplasma SAT → Brucellosis MAT → Leptospirosis

02:06:42

Bacterial Vaginosis

Caused by:

- Gardenerella
- Mobilincus
- Provetella
- Amsel's Criteria:
  - → ph > 4.5
  - → Discharge + 10%KOH → FISHY Odour
    - ↓
    - WHIFF's Test

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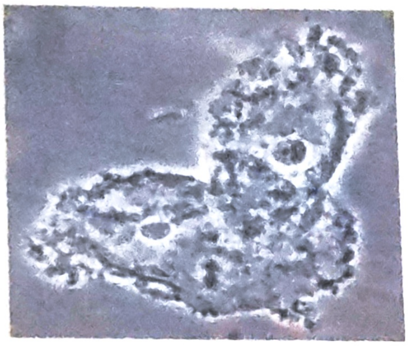
PrepLadder

→ Clue cells: epithelial cells with bacteria

Nugen's Criteria:

→ For Bacterial Vaginosis

→ Grading of replacement of normal Flora of vagina by Pathogenic Organisms



Clue cells

Bacteroides 02:10:03

- GNB
- Anaerobe obligate
- B. fragilis → Normal commensal of gut

↓

May lead to infection in those who undergo abdominal or gut Surgery

- a/w 'Brain abscess'
- Foul smelling pus → anerobic growth
- LPS
  - Endo toxic action
  - Mild potent endotoxin
  - May lead to 'Shock & DIC'

MISCELLANEOUS

Streptobacillus moniliformis

- Causes HAVERHILL fever
- a/w Rodents (rat bite fever)

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PrepL

SPIRILLUM MINUS → causes Rat Bite Fever

HUMAN BITE

- a/w Eikenellacordens
- a/w anaerobic Streptococci

CAT & DOG BITE

a/w Pastuerella multocida

SCROMBOID FISH POISONING

E. coli › morgenella

Morgenella › proteus › E.coli (as per WHO)

Non Sporing Anaerobes

Gm +ve

Engineers	Eubacterium
Loe	Lactobacillus
M	Moblincus
B	Bifido bacterium
A	Actinomyces
Program	Propionibacterium acnes

Gm –ve

B Bacterioids

P Prevotella, Porphyromonas

L Leptotrichia

Fuse Fusopacterium

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Unit: 3 MYCOLOGY

Chapter 8

MYCOLOGY

FUNGI → EUKARYOTIC Protista

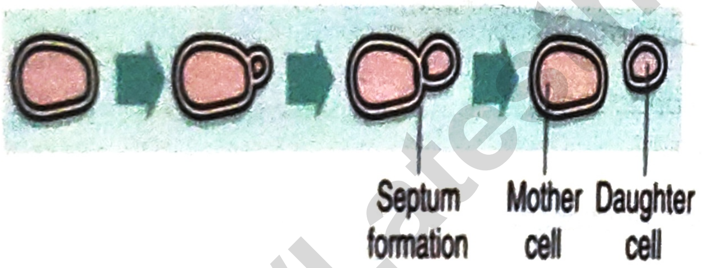
CELL WALLS → Chitin, Mannan & Polysaccharides

Cytoplasmic membrane → Sterols

Possess True Nuclei, Nuclear membrane & paired Chromosomes

SIMPLEST TYPE OF FUNGUS → Unicellular Budding yeast

* MYCOPLASMA Cytoplasmic membranes are also made up of Sterols



HYPHAE → Elongated tubular structure

MYCELIUM → Tangled mass of hyphae

HYPHAE → Septate or Aseptate

MYCELIUM
→VEGETATIVE → Grows into medium
→AERIAL → projects from surface

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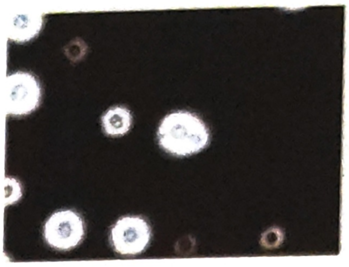
Prep

CLASSIFICATION

DEPENDING ON CELL MORPHOLOGY

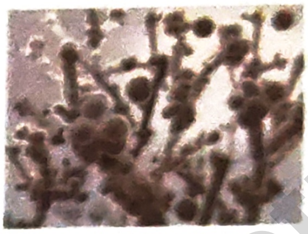
00:02:42

1. YEAST / TRUE YEAST
   - →Spherical or Ellipsoidal cells
   - →Reproduce by Budding
   - →Eg: Cryptococcus neoformans



YEAST

2. YEAST LIKE
   - Partly as yeast
   - Partly as elongated cells: PSEUDOHYPHAE
   - Eg: Candida Albicans
     - can show True hyphae / Pseudohyphae
     - can show yeast/yeast like [main form]



YEAST LIKE

3. MOULDS / MOLDS
   - True mycelia
   - Reproduce by Spores
   - Eg: Rhizopus, Aspergillus

MOULDS

4. DIMORPHIC FUNGI
   - In host tissue or cultures at 37°C → Yeasts
   - In soil & cultures at 22°C/25°C → Moulds/Molds
   - Eg:

H → Histoplasmosis	C → Coccididomycosis
S → Sporotrichosis	P² → Para coccididomycosis
B → Blastomycosis	→ Penicilliosis

Based on Sexual Spore formation

00:08:00

1. PHYCOMYCETES / ZYGOMYCETES
   - Non - Septate hyphae
   - Obtuse angled
   - Forms asexual Spores : Sporangia
   - Sexual Spores → Zygospores

PrepLadder

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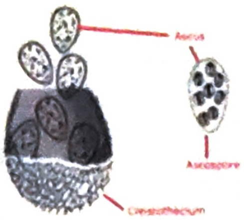
2. ASCOMYCETES

- Sexual spores → Ascospores in an Ascus



Ascospores in an ascus

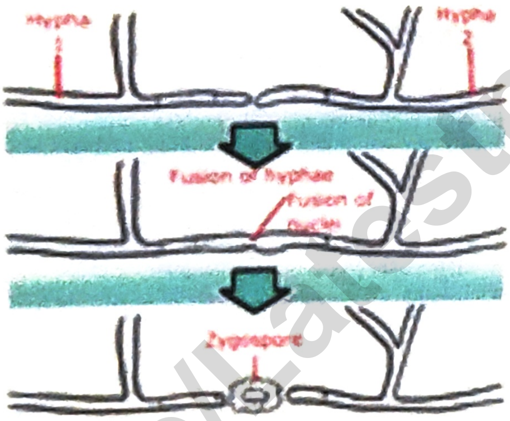
3. BASIDIOMYCETES

- Sexual spores on a basidium
- Seen in Cryptococcus neoformans

4. FUNGI IMPERFECTI [DEUTEROMYCETES]

- No sexual phases

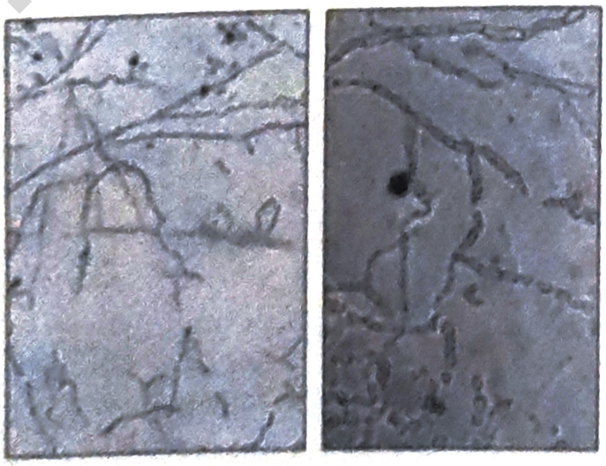
Sexual spores		Asexual spores	
Z →	Zygospore	A →	Arthrospore
A →	Ascospore	B →	Blastospore
B →	Basidiospore	C →	Chlamydospore



LAB DIAGNOSIS

By Microscopic examination

1. Wet mount with 10% KOH → can see fungal elements



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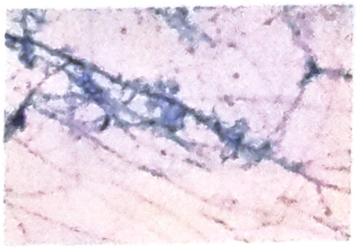
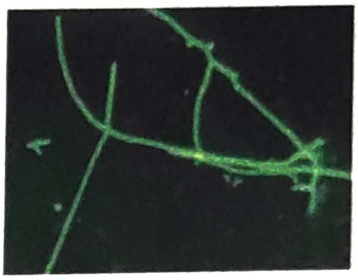
2. Tease mount with Lactophenol cotton blue [LPCB]



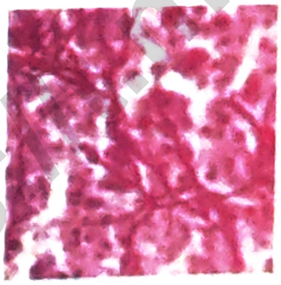
3. Calcoflour white staining [Fungal Fluorescent staining]



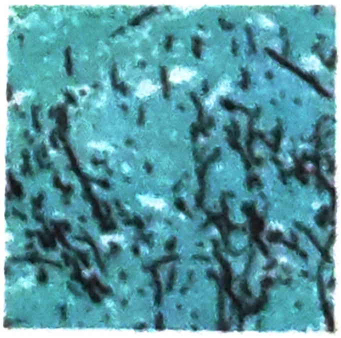
4. Slide culture

Slide culture

5. Periodic Acid Schiff [PAS]



6. Gomori Methanamine Silver stain
- Used For Pneumocystis carnii/Jiroveci
- Stains green
- Trophozoites of Pneumocystis carnii are stained green but Cysts are stained black (as they have glycogen)



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PrepLadder

- Pnemocystis Carnii
  - causes Pneumocystis carnii pneumonia in HIV +ve with CD_4 count < 200
  - DOC → Cotrimoxazole

CULTURE MEDIA

00:16:17

1. SABOURAUD'S DEXTROSE AGAR [SDA] [pH = 5.6 (5.4-5.6)]
   - Universal fungal culture media
2. CZAPEK - DOX MEDIUM
3. CORN MEAL AGAR

SDA

- Cycloheximide → Selective agent → makes media selective for fungus
- Incubation Time & Temperature
  - at room temp. (22°C) → Weeks
  - at 37°C → 4 days

INDENTIFICATION

00:17:19

1. By Colony & Fungal morphology
2. Rapidity of growth, color
3. Morphology of the colony → Obverse
4. Pigmentation → Reverse

TEASE MOUNT OR SLIDE CULTURE

→ Hyphal diameter, septa & special structures are important

SPECIAL HYPHAE

1. Spiral hyphae → Spring like helical coils
2. Racquet hyphae → Resembling Tennis Racquets
3. Favic chandelier → Numerous short branches appearing at the ends of hyphae

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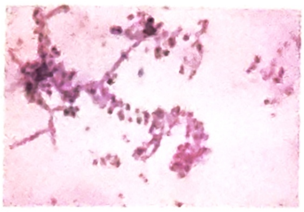
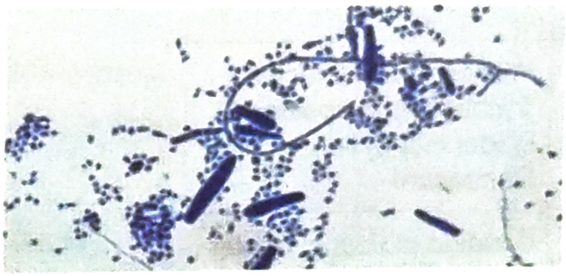
Prep



Spiral Hyphae   Racquet Hyphae   Favic Chandelier

CONIDIA

1. Microconidia → Small, single celled
2. Macroconidia → Large, single or multi celled



SPORES

1. Blastospores → Budding
2. Chlamydospores → Thick walled resting spores formed by rounding up & thickening of hyphae
   → Eg: Characteristically seen in Candida albicans on Corn meal agar
3. Arthrospores → Along the hyphae by Segmentation & Condensation
   → Rectangular
   → Eg: Coccididomycosis

Classification Based On Location   00:20:53

Mycoses

1. Superficial Mycoses → Superficial Layer Of Skin Involved
   Subcutaneous Mycoses → Skin & Subcutaneous tissue involved
2. Deep Mycoses/Systemic Mycoses
3. Opportunistic Mycoses

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PrepLadder

CLINICAL CLASSIFICATION OF MYCOTIC INFECTIONS

Area of Predominant involvement	MYCOSIS	ETIOLOGY
1. SUPERFICIAL	Pityriasis versicolor	Malassezia furfur Malassezia globosa (skin)
	Tinea nigra	Phaeoannellomyces werneckii / Exophila werneckii
	White piedra	Trichosporon beigelii
	Black piedra	Piedra hortae
2. CUTANEOUS	Dermatophytosis	Microsporum species, Trichophyton species & Epidermophyton Floccosum
	Candidiasis of skin, mucosa or nails	Candida albicans, other Candida species
3. SUB CUTANEOUS	Sporotrichosis	Sporothrix Schenckii
	Chromomycosis	Philaphora verrucosa, Fonsecaea pedrosoi & others
	Mycetoma	Pseudallescheria boydii, Madurella mycetomatis & others
	Mnemonic M-Mycetoma R-Rhinosporidiosis C-Chromomycosis S-Sporotrichosis	
4. SYSTEMIC MYCOSES	(HSBC P^2)	
Primary Mycoses	Coccidioidomycosis	Coccidioides Immitis
	Histoplasmosis	Histoplasma capsulatum
	Blastomycosis	Blastomyces dermatidis
	Paracoccidiomycosis	Paracoccidioides brasiliensis

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Opportunistic Mycoses	Candioidomycosis	Candida albicans, other Candida species
	Cryptococcosis	Cryptococcus neoformans
	Aspergillosis	Aspergillus fumigatus, other Aspergillus sp.
	Mucormycosis	Species of Rhizopus, Absidia, Mucor & others

SUPERFICIAL MYCOSES

00:26:53

Include

A. Surface infections

B. Cutaneous Infections

SURFACE INFECTIONS

TINEA VERSICOLOR

00:29:20

- Caused by Malassezia furfur
- Presents with discrete hyperpigmented or hypopigmented macules / scales on chest or Upper back
- COLONY → Yeast like consistency
- Microscopy → Spherical, oval or cylindrical cells

CLINICAL FEATURES

1. HIGHER incidence in tropics
2. PREDISPOSING FACTORS

→Perspiration

→Corticosteroids

→Malnutrition

→Heredity

→Immunosuppression

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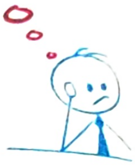
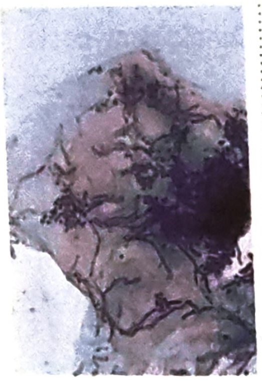


DIAGNOSIS



1. Microscopic Examination
   - Skin Scrapings

   →Scales + 10% KOH→ SPAGHETTI & MEATBALL APPEARANCE

   →Scales + Wood's lamp Examination

   → Yellow Fluorescence

   →Scales + SDA & Olive oil overlay [Lipophilic fungus]→FRIED EGG COLONIES

TREATMENT

Topical applications of

1. Selenium sulphide 10 min daily for 7 days for 1 year
2. Miconazole, Ketoconazole

2. TINEA NIGRA 00:34:07

- Superficial, chronic & asymptomatic infection of Stratum corneum
- Caused by Cladosporum wernickii or Exophila wernickii
- Saprophyte & dematiaceous [pigmented hyphae]
- Microscopically
  - Budding yeasts & chains of yeast cells
  - Pigmented hyphae

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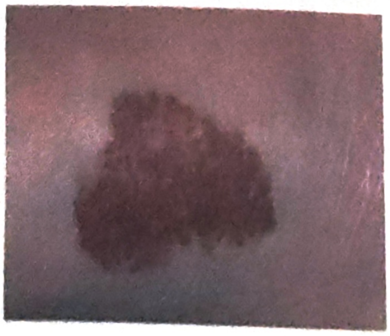
- Lesions
  - Solitary, innocuous macule with brownish color at the advancing periphery
  - Found in palms, sole, face & finger



TINEA NIGRA

- Diagnosis
  - By direct KOH & culture of skin scrapings from periphery of the lesion

- TREATMENT
  - Topical Keratolytic solution of Sulphur Salicylic Acid or Tincture of Iodine

PIEDRA

00:37:07

- Fungal infection of hair by appearance of firm, irregular nodules along the hair shaft
- Black Piedra → Piedra hortae
- White Piedra → Trichosporon beigelli

- TREATMENT
  - Removal Of Hair & Application of Topical Antifungal Agent

WHITE PIEDRA

BLACK PIEDRA

DERMATOPHYTES / CUTANEOUS INFECTIONS

- Group of filamentous Fungi that infect only superficial Keratinized tissues - skin, hair & nails
- Clinical condition → Tinea or Ringworm
- They secrete : Keratinases, Proteolytic enzyme that digest Keratin of hair, nails & epidermis
- Attack rate is higher in institutions & overcrowded population

CLASSIFICATION

3 GENERA

1. Trichophyton → Skin, hair, nails
2. Microsporum → Skin, hair
3. Epidemorphyton → Skin, nails

MORPHOLOGY → Identification by Colony appearance & Microscopy

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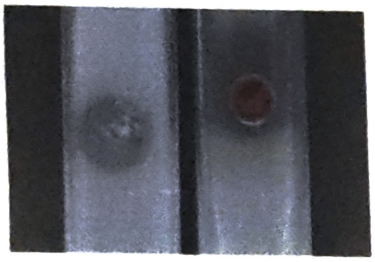
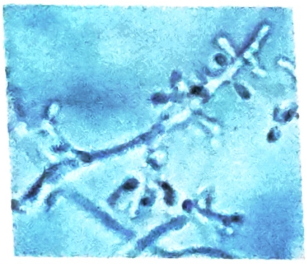
PrepLadder

TRICHOPHYTON

→ Cylindrical / Pencil shaped, smooth walled Macroconidia & numerous Microconidia



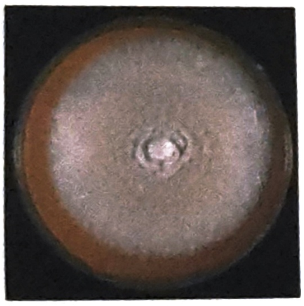
T.RUBRUM

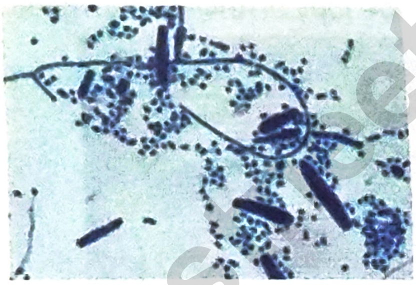


T.RUBRUM MICROCONIDIA

T.RUBRUM MACROCONIDIA

TRICHOPHYTON MENTAGROPHYTES

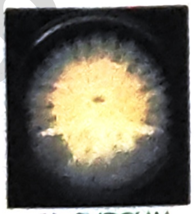




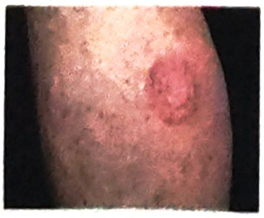
TINEA VIOLACEUM

MICROSPORUM

- Multicellular Macroconidia [Spindle shaped] with rough walls & few Microconidia



M.GYPSUM



M.canis

SPINDLE SHAPED MACRO CONIDIA OF M.GYPSUM

SPINDLE SHAPED MACRO CONIDIA OF M.CANIS

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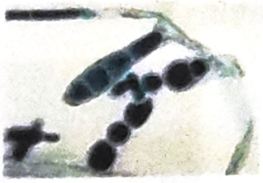
EPIDERMOPHYTON

- Only Macroconidia which are smooth walled & Clavate / Club shaped / Boat shaped

EPIDERMOPHYTON


EPIDERMOPHYTON MACROCONIDIA

PATHOGENSIS & IMMUNITY

- By contact with dermatophytes in soil, animals or by direct contact or fomites
- Host susceptibility is increased by : Moisture, warmth, genetic predisposition & skin composition of sebum

EPIDEMIOLOGY

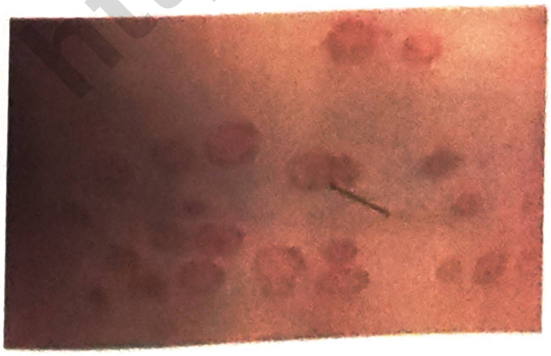
- Anthropophilic
- Zoophilic
- Geophilic

CLINICAL FEATURES

- Erroneously termed : Ringworm or Tinea because of raised circular lesion

TINEA CORPORIS

- Lesion is annular, sharply marginated with raised border
- This pustular well circumscribed, elevated, crusted lesion is called as MAJOCCHI'S Granuloma
- Caused by T. rubrum



Majocchi's Granuloma

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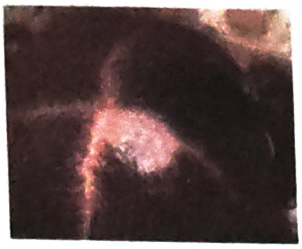
TINEA CAPITIS

- Infection of the shaft of hairs of scalp
- May lead to Alopecia & scarring



INFLAMMATORY → KERION, FAVUS

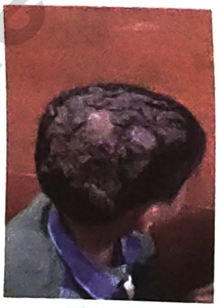
a. KERION (HONEY COMB)

- Painful boggy swelling over the scalp
- Painful severe form of inflammatory reaction
- Caused by Trichophyton verrucosum, T. mentagrophytes

KERION

b. FAVUS

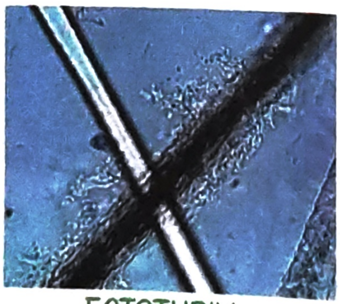
- Dense crust formation over the scalp
- Caused by Trichophyton Schoenleinii



FAVUS

ECTOTHRIX

- Sheaths of spores around the shaft
- Caused by Microsporum canis, M. audonii



ECTOTHRIX

ENDOTHRIX

- Arthrocondia within the hair shaft
- Caused by

  T → Trichophyton tonsurans

  V → Trichophyton violacaeum

  S → Trichophyton schoenleinii

ENDOTHRIX

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LAB DIAGNOSIS

- Skin Scrapings
  Nail Scrapings
  Infected Hair } +10% KOH → Fungal Elements are observed

- Direct Examination under Wood's LAMP
  - Tinea capitis → Infected hair exhibits greenish Fluorescence

TINEA BARBAE [BARBER'S ITCH] → Infection of beard area of the face

TINEA CRURIS/DHOBI ITCH / JOCK ITCH

- Infection of groin → seen in men
- Involves Perineum, Scrotum, Perianal regions

TINEA MANUUM 00:52:43

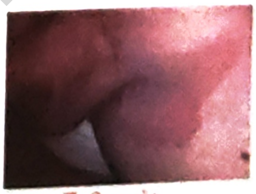
- Infection of skin of hands
- Diffuse Keratosis of Palms & fingers by E. Floccosum, T. rubrum, T. mentagrophytes

TINEA UNGUIUM → Infection of nail plates at the free edge of the nail

TINEA PEDIS [ATHLETE'S FOOT]

- Toe web scaling, fissuring, maceration
- Also Known as Sandal ringworm

T.Barbae


T.Cruris

T.Mannum

T.Pedis

T.Unguium

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ID REACTION

- Allergic response to fungal antigens
- Infection in feet elicits the allergic reaction on the hands [always away from site]
- Eg: Tinea pedis elicits allergic reaction on the hands

LAB DIAGNOSIS

A. Skin Scrapings, Nail Clippings, Infected Hair } + 10% KOH → FUNGAL ELEMENTS OBSERVED

B. Skin scrapings, infected nails, hairs + SDA /DIM [Dermatophyte Identification media]

↓

Culture growth [may take 2-3 days /weeks/months]

C. Culture Growth + LPCB mount & Observe

	Macro conidia	Micro conidia
Trichophyton	→ Cylinderical/Pencil shaped	→ Numerous
Microsporum	→ Spindle Shaped	→ Less
Epidermophyton	→ Club / Boat Shaped	→ Absent

SUB CUTANEOUS MYCOSIS

00:59:19

- Involves skin & sub cutaneous tissues
- Eg: M → Mycetoma
  - R → Rhinosporidiosis
  - M → Chromoblastomycosis / Chromomycosis
  - S → Sporotrichosis [dimorphic]

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MYCETOMA / MADURA FOOT

ACTINOMYCETOMA	EUMYCETOMA
→ Bacterial	→ Fungal
→ Numerous Sinus & Swelling	→ Single Sinus & Swelling
→ Purulent discharge	→ Serous discharge
→ White / Yellow granules • Actinomadura madurae • Nocardia caviae • Nocardia asteroides • Nocardia brasilensis → Red granules • Actinomadurae pelletri	→ Black granules • Madurella grisea • Madurella mycetomatis → White granules • Acremonium • Pseudoallescheria boydii
→ Crush granules & see under microscope → <2 μ bacilli	→ Crush granules & see under microscope → >2 μ hyphae / >2 μ Fungal colonies
→ On X ray → Osteolytic lesions	→ On X ray → Osterosclerotic lesions
→ Responds well to Antibiotics	→ Responds very poorly to Anti fungals [amputation may be required]

Mycetoma

Madura foot

BOTRYOMYCOSIS → Mycetoma like lesion
→ Mainly caused by Staph. aureus

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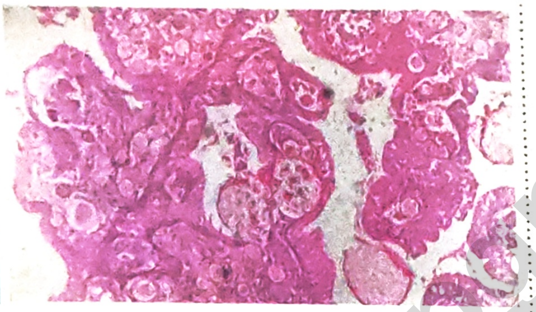
RHINOSPORIDIOSIS

- Caused by Rhinosporidium seeberi [AQUATIC PROTOZOA]
- Present with U/L or B/L Nasal polyps [MC presentation]
- POLYP BIOPSY → Shows Spherule with endospores
- Rx → Sx Polypectomy [Best]
- Non cultured on SDA

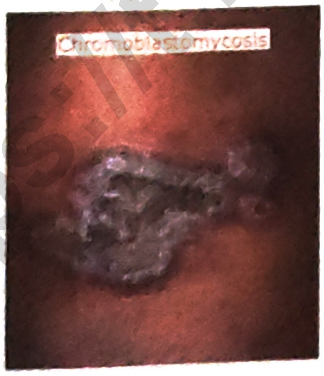


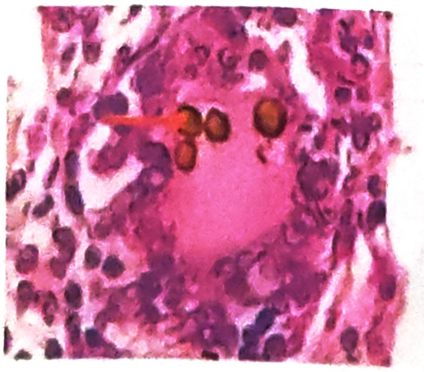
CHROMOBLASTOMYCOSIS / DEMATIACEOUS FUNGUS / PIGMENTED FUNGUS

- Involves skin & sub cutaneous tissues
- THORN PRICK / WOOD PRICK

  ↓

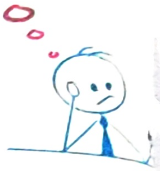
  VERRUCCOUS LESION / WARTY LESION
- BIOPSY OF WARTY LESION + H & E SHOWS
  - Brown colored Globose bodies
  - SCLEROTIC BODIES / MEDLAR BODIES
  - Show Copper Penny Appearance





MEDLAR BODIES

Examples →
1. Alternaria
2. Bipolaris
3. Curvularia
4. Phialophora
5. Cladosporium
6. Exophila



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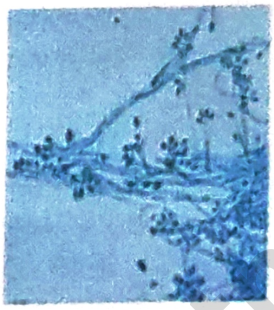
SPOROTRICHOSIS / ROSE GARDENER'S DISEASE

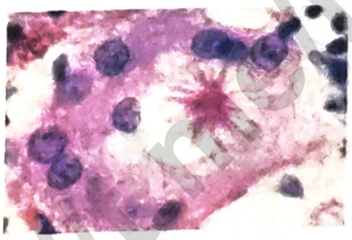
- Caused by Sporothrix schenkii
- Spread by thorn Prick [Gardener's disease]
- ↑ ed incidence in & around Valentine's day
- Lymphocutaneous spread is seen
- Dimorphic fungus [at 22-25°C → Moulds, at 37°C → Yeast]
  - Mold → Flower like sporulation
  - Yeast → Cigar shaped yeast cells or
    → Narrow based budding yeast cells

SPOROTRICHOSIS



Sporothrix schenkii

→ ASTEROID BODIES are seen
- Basophilic material is surrounded by Eosinophilic material → Splendore - Hoepelli Phenomenon



Asteroid Body

DEEP MYCOSIS

01:25:22

Dimorphic Fungus Cause DEEP MYCOSIS Except Sporotrichosis

HISTOPLASMOSIS [DARLING'S DISEASE]

- Systemic, granulomatous, intracellular infection
- Affects Reticuloendothelial system
- Caused by Histoplasma capsulatum [Dimorphic Fungi]
- Source → Soil droppings of Pigeons & Bats. Fungus is found in these droppings

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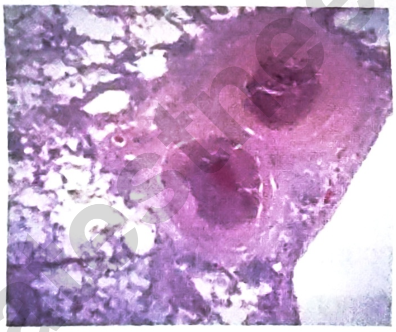
- Mode of Transmission → Aerosol transmission in form of Multicellular filamentous mycelia
- Incubation Period (IP) = 10-16 days
- Lung involvement → Blood → Viscera

CLINICAL FEATURES

1. PULMONARY DISEASE

Acute Form	Chronic Form
- Influenza like disease - Resolves within 2-3 weeks - Shows lymphocyte rich infiltrate in lungs	- Haemoptysis - Apical cavities - Calcification } resembles TB



2. CUTANEOUS & SUB CUTANEOUS DISEASE → Petechial lesions

3. DISSEMINATED DISEASE

- Anemia
- Leucopenia
- Hepatosplenomegaly
- Multiple lymphadenopathy

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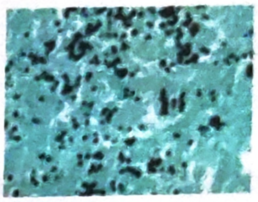
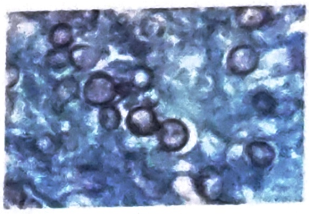
LAB DIAGNOSIS

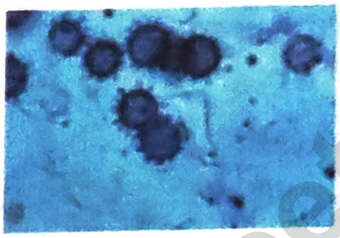
1. Sputum
2. Sternal Bone marrow
3. Lymph node

MICROSCOPY





- Giemsa Stain
- Gram's Stain
- Shows - Intracellular, Oval, Budding Yeast cells



CULTURE

- SDA with Actidione
  - White Cottony mycelial growth with Tuberculate spores / Macroconidia [Mold form]
- Potato dextrose agar

SEROLOGY

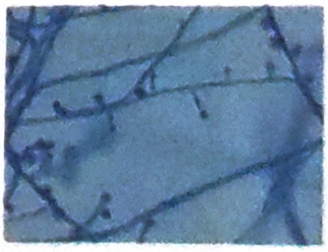
- Immunodiffusion
- Latex agglutination
- Complement Fixation test
- Histoplasmin Skin test - DTH [delayed type Hypersensitivity]

MOLD → TUBERCULATE MACROCONIDIA

YEAST → NARROW BASED BUDDING YEAST CELL

narrow base yeast cell

BLASTOMYCOSIS / NORTH AMERICAN BLASTOMYCOSIS / CHICAGO DISEASE / GILCHRIST DISEASE



- Caused by Blastomyces dermatidis
- Infection by inhalation of Conidia
- Pyo - granulomatous Fungal infection of Respiratory tract
- Dissemination to skin, bone & genito urinary system

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PrepLadder

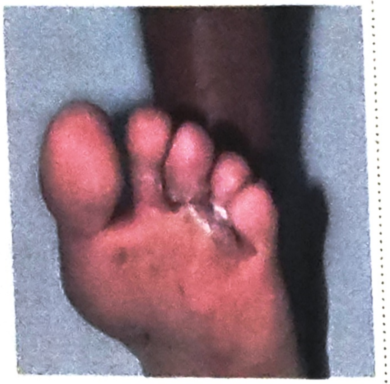
CLINICAL FEATURES

1. CUTANEOUS BLASTOMYCOSIS

- Commonest Form
- Trauma
- Papule → Secondary nodules

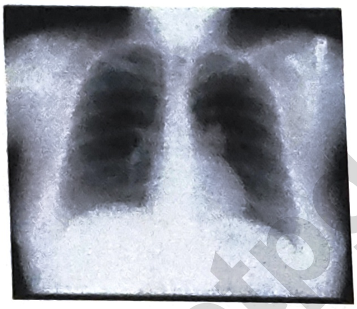
↓

Coalesce to form Ulcers



2. PULMONARY BLASTOMYCOSIS

- Resemble Pulmonary TB, Carcinoma



3. DISSEMINATED BLASTOMYCOSIS

- Hematogenous spread to
  - Bones
  - Genitourinary tract

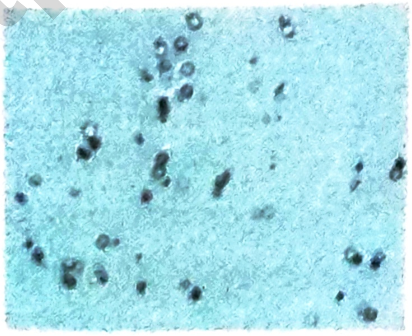
LAB DIAGNOSIS

SAMPLES

- PUS
- Sputum
- Tissues
- Bronchoalveolar lavage [BAL] fluid

1. Microscopy

- GMS Staining → Multiple budding yeast
- Calcofluor White



GMS stain

Calcofluor White

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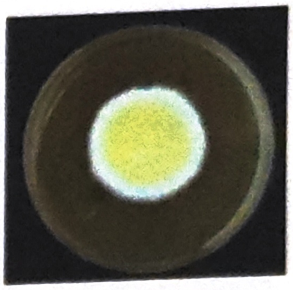
Prep

2. CULTURE



8 Broad Base yeast cell

- SDA at 25°C
  - Fluffy White to Tan colored
  - Fine branched Filaments with Conidia on terminals
- On BHI at 37°C
  - Waxy & heaped mass

YEAST FORM	→	Broad Based budding Yeast cell
[at 37°C]	→	Figure of 8 appearance

COCCIDIOIDOMYCOSIS

- Caused by Coccidioides immitis
- Pathogenic dimorphic fungi
- In culture, it produces Arthrocondia within the mycelia at 25°C
- In tissue, it produces Spherules with endospores at 37°C
- Found in soil

↓

Grows as a mold under dry conditions

↓

Arthroconidia break out of mycelia

↓

Become airbone

- Incubation Period → 1-4 weeks

CLINICAL FEATURES

1. PULMONARY INFECTIONS : Primary type

- San Joaquin valley fever or Desert Rheumatism
- d/t Inhalation of Conidiae
- Acute influenza like illness → Self limiting
- Bronchopneumonia happens rarely

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2. Primary Cutaneous Type

- Nodule → Ulceration [DD - Syphilis, TB ulcer]
- Lymphangitis
- Lymphadenopathy

3. DISSEMINATED TYPE → Rare

- Fatal if occurs [Granulomas in Internal organs]
- Meninges, bones, joints are involved

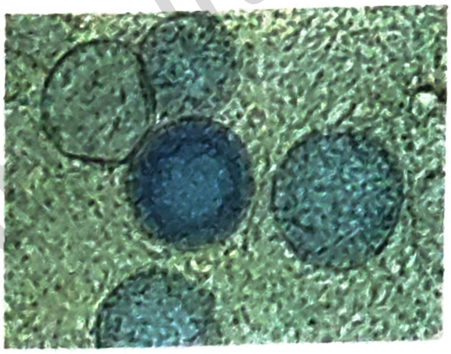
LAB DIAGNOSIS

SAMPLES

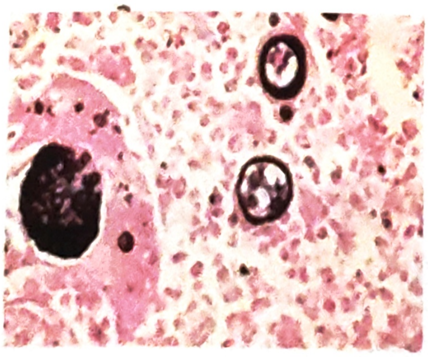
- Skin scrapings
- Sputum
- Bronchial washings
- CSF
- Pleural Fluid & Blood etc

1. DIRECT MICROSCOPY OF SKIN SCRAPINGS

- 10% KOH & Parker ink
- Sporangia of Coccidioides immitis
- It is diagnostic
- TISSUE SECTION → Spherules of Coccidioides immitis with endospores



Direct Microscopy



Tissue Section

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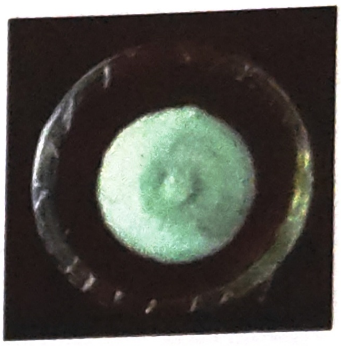
2. CULTURE

- SDA [at 25°C]
- Brain Heart Infusion agar with 5% sheep Blood
  - Colonies are initially moist & glabrous but rapidly becomes Greyish White in color with a Tan reverse

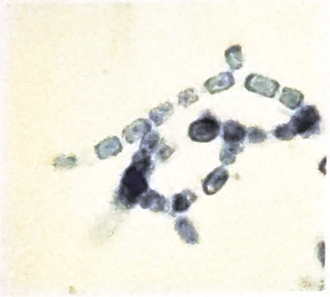


MICROSCOPY MORPHOLOGY

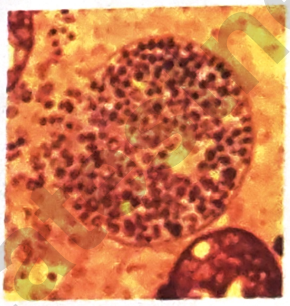
- Single celled, Hyaline
- Rectangular to barrel shaped, alternate Arthroconidia

At 25°C → Arthroconidial arrangement [MOLD]

At 37°C → Spherule with endospore [Yeast]



Arthroconidial arrangement



Spherule with endospore

3. SKIN TEST → Coccidioidin

4. SEROLOGICAL TESTS

- Immunodiffusion test
- Complement Fixation test

PARACOCCIDIOIDOMYCOSIS / SOUTH AMERICAN DISEASE / LUTZ MYCOSES 01:45:55

- Caused by Paracoccidioides brasiliensis
- Dimorphic Fungi
- Common in coffee growing belt
- Favourable conditions
  - ↑ Humidity
  - ↓ Temperature [23°C]

https://t.me/latestneetpgnotes

PrepLadder

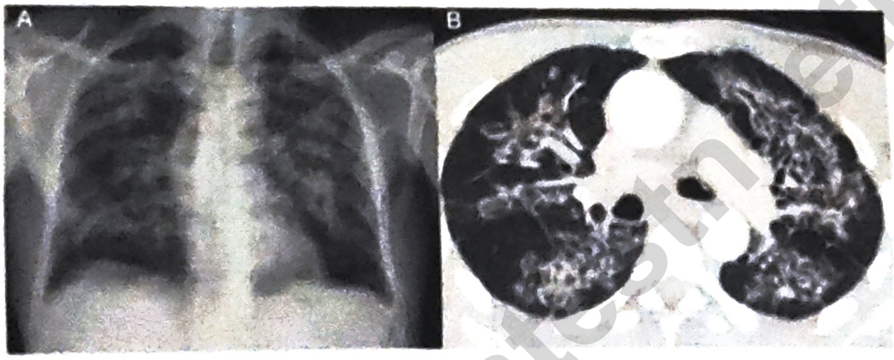
CLINICAL FEATURES

1. MUCOCUTANEOUS FORM → Ulcerated lesion

2. LYMPHATIC FORM → Cervical lymphadenopathy, Bull neck appearance

3. VISCERAL FORM

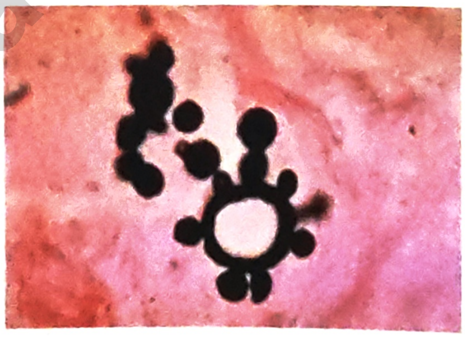
- Spreads by Lymphatic or Hematogenous route
- Lung (90% of cases)
- CNS, Bones
- B/L Patchy Pneumonia with Nodular Miliary shadow



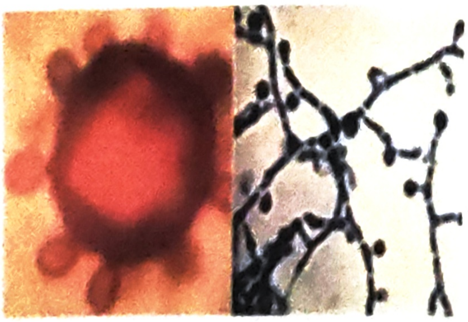
LAB DIAGNOSIS

SAMPLES

- Sputum
- Bronchoalveolar lavage
- PUS



1. MICROSCOPY → Multipolar budding yeast cells
2. CULTURE
   - YEAST → Mickey mouse appearance / Mariner's Wheel appearance / Pilot wheel appearance
   - MOLD → Mycelial form



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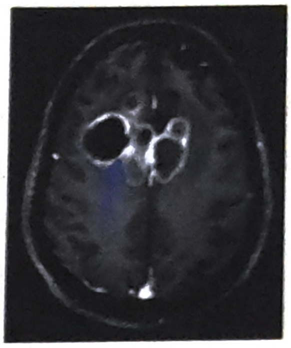
PrepLadder

3. SEROLOGY

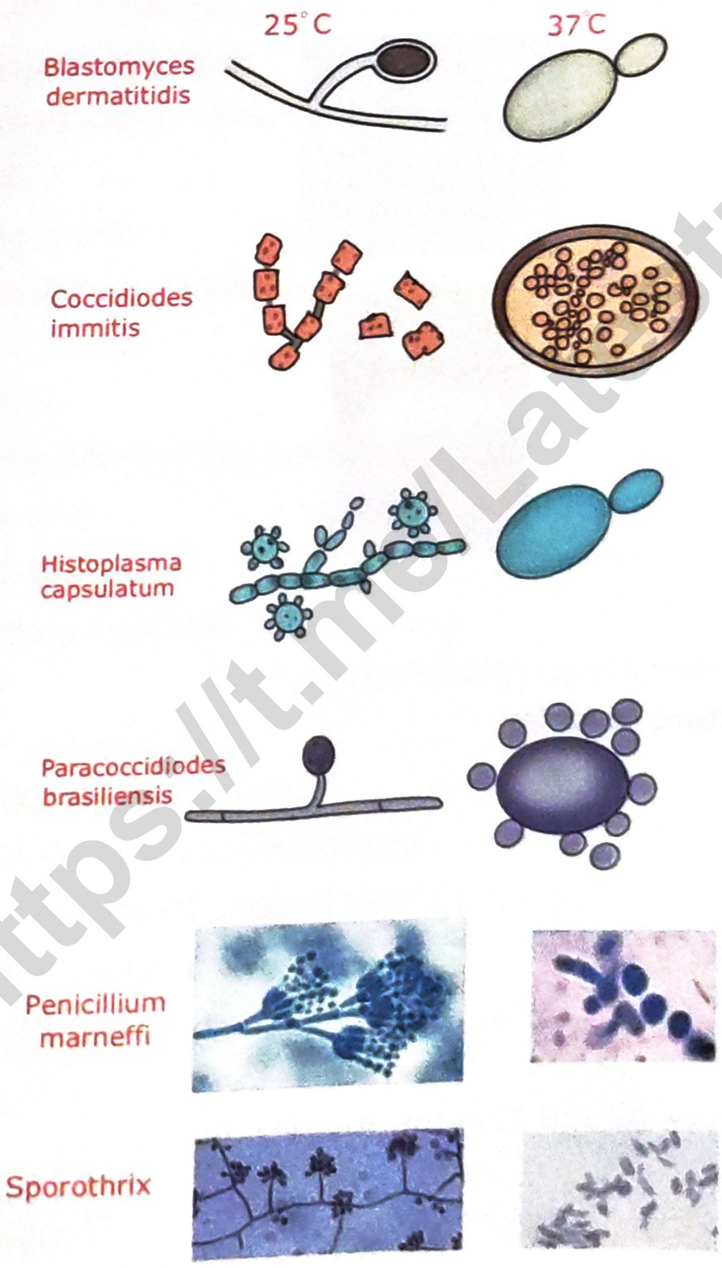
- Antibody detection
- Latex agglutination test
- Complement Fixation test
- Immunodiffusion



4. RADIODIAGNOSIS

- CT Scan
- MRI

Dimorphic fungi



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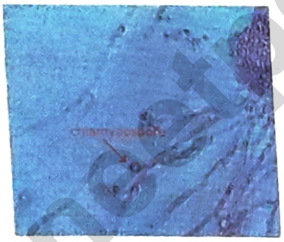
CANDIDA [YEAST LIKE] 01:51:42

CANDIDA ALBICANS

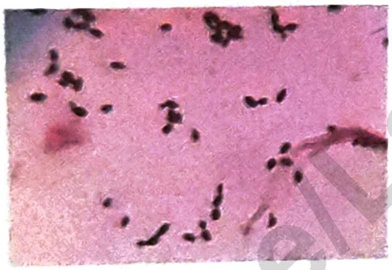
- Shows
  1. REYNOLD BRAUDE PHENOMENON [Germ tube Formation]
  2. CHLAMYDOSPORE FORMATION ON CORN MEAL AGAR

- REYNOLD BRAUDE PHENOMENON [Culture growth + Human plasma]
  - Shows germ tube formation [GERM TUBE TEST]
  - On corn meal agar (nutrient deficient medium), Chlamydospores are formed [at 20°C]

REYNOLD BRAUDE PHENOMENON



Oral thrush



CANDIDA BUDDING

NON - CANDIDA ALBICANS [NCA]

- C.Krusei → Naturally resistant to Azoles
- C.Kefyr
- C.glabrata
- C.stellatoidea
- C.guillermondii
- C.tropicalis
- C.vishwanthi
- C. aureus → Causes worldwide infections mostly in Immunodeficient

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Prep

CANDIDA ALBICANS - DISEASES

1. ORAL THRUSH
2. OESOPHAGEAL CANDIDIASIS
3. VAGINAL CANDIDIASIS
4. CANDIDAEMIA [Candida yeast cells in blood]
5. CANDIDURIA [Candida Yeast Cells In Urine]
6. ONYCHOMYCOSIS [Nail Folds are involved]
7. UTI/RTI
8. MENINGITIS

CANDIDA INFECTIONS ARE COMMON IN

- Neutropenic individuals
- Conditions with ↓ Cell Mediated Immunity [CMI]
  - AIDS
  - Malignancies
  - Chronic Muco-cutaneous Candidiasis [T-cell disorder]

LAB DIAGNOSIS

- Take Tissue Biopsy; Samples & do
  - Gram stain → Yeast cells are seen
  - Cx on SDA → Creamy, pasty colonies
  - Chromagar method → Colourful agar
    → Used for CA/ NCA identification

* CA → Candida albicans

* NCA → Non Candida albicans

- SUGAR FERMENTATION METHODS
- β GLUCAN TESTING FOR INVASIVE CANDIDIASIS
- PCR FOR Candida species

CHROMAGAR

SDA

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PrepLadder

CRYTOCOCCUS NEOFORMANS

02:05:42

- Present in Pigeon faeces

  ↓

  Aerosol transmission

  ↓

  Lung Involvement

  ↓

  Haematogenous spread

  ↓

  MENINGES INVOLVED [Meningitis]

  } NEUROTROPISM [For HIV +ve individuals]

- MC cause of Meningitis in HIV +ve → Cryptococcus neoformans

LAB DIAGNOSIS

INDIA INK STAINING

SDA

1. CSF SAMPLE

   a. Indian Ink staining

   - Negative staining
   - Organisms illuminate against dark background

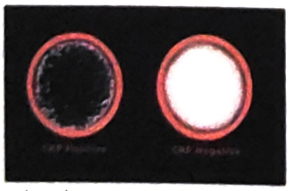
   b. Culture

   - On SDA → Mucoid, smooth
   - On Niger seed agar → Blackish Brownish colonies

UREA HYROLYSIS TEST POS
FOR CRYPTOCOCCUS

2. LATEX CARD AGGLUTINATION TEST for Cryptococcal Antigens

3. PCR for Cryptococcal neoformans



Latex card test

* DOC For Cryptococcal Meningitis → Amphotericin B + Flucytosine

ZYGOMYCETES

02:12:01

- Rhizopus
- Mucor
- Absidia

} Cause infections in Terminally ill patients

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PrepLadder

RHINO CEREBRAL ZYGOMYCOSIS

- Caused by Rhizopus species
- Fulminant disease
- Tissue Biopsy + H & E Staining → Aseptate hyphae, Obtuse angled branching

- Tissue Biopsy + SDA

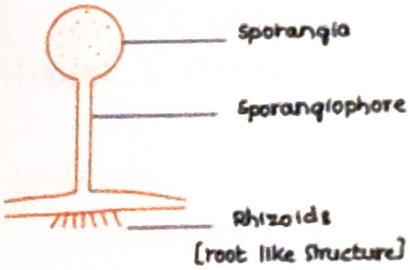
↓

Culture growth

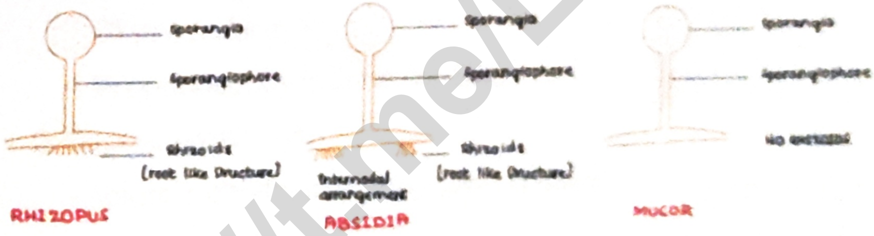
↓

LPCB mount [Lactophenol cotton blue mount]

↓



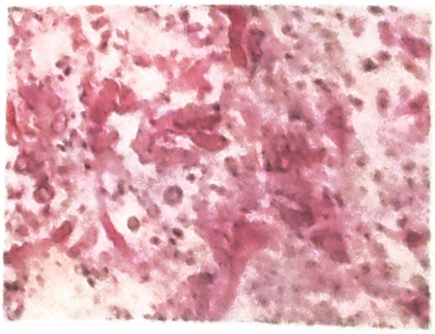
DIFFERENT ZYGOMYCETES



MUCOR MYCOSIS

02:17:28

- Predisposes in
  - DM
  - Frequent blood transmission
  - Chronic dialysis



Mucormycosis

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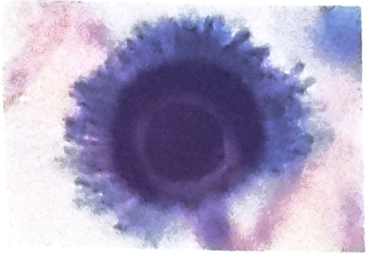
ASPERGILLUS

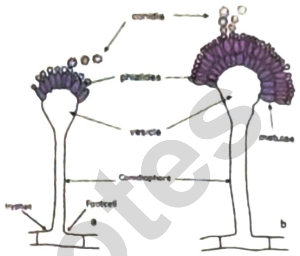
02:19:00

A. fumigatus → Causes

- ABPA (Allergic Broncho Pulmonary Aspergillosis)
- Aspergilloma (Fungal ball)
- Oculomycosis
- Otomycosis



ASPERGILLUS



STRUCTURE OF ASPERGILLUS

A. niger

- Causes Otomycosis
- Blackish crust is seen

A. Clavatus

- Causes Maltster's Lung
- Common in Barley / Malt factory workers
- Occupational disease

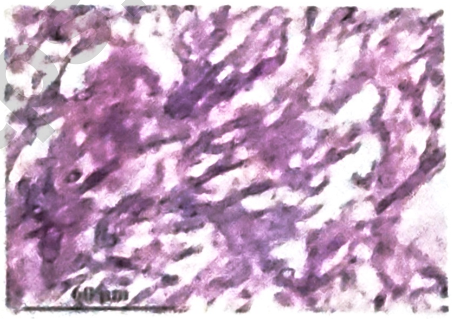
A. flavus

- Produces Aflatoxin
- Aflatoxin is a/w HCC (Hepato cellular carcinoma)

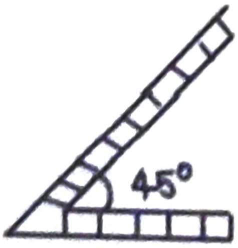
LAB DIAGNOSIS

Tissue Biopsy + H & E Staining

- Septate hyphae
- Dichotomous branching
- Acute angled (mostly 45°C)



Aspergillus Dichotomous branching



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PrepLadder

ERGOTOXICOSIS [ERGOT POISONING] 02:24:38

- Edibles + Claviceps purpurea seeds → Ergotoxicosis
- Consumption of contaminated edibles leads to
  1. St. Vitus Dance &
  2. St. Anthony's fire
- Gastric irritation occurs

  ↓

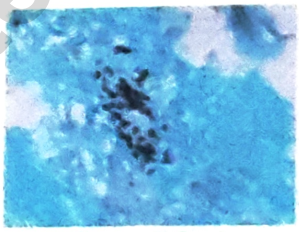
  Restlessness [St. vitus Dance]

PNEUMOCYSTIS CARNII / PNEUMOCYSTIS JIROVECII

- Protozoa → Cyst/Trophozoite stage present which favours it to be Protozoa
- Fungus → Chromosomal studies favours it to be Fungus

- Pneumocystis carnii pneumonia
  - Occurs in persons with CD_4 counts → < 200 [HIV +ve]
  - DOC → COTRIMOXAZOLE
  - LAB DIAGNOSIS

1. BAL [Broncho Alveolar Lavage] Fluid
   - OT procedure
   - Bronchoscopy is done for BAL
2. Gastric washings
   - Taken in children as they tend to swallow their sputum



P.carnii Cyst

BAL / GASTRIC WASHINGS

↓

GMS staining [Gomorii Methanamine Silver]

↓

Trophozoites → Green (GMS is naturally green colored)

Cysts → Black (d/t Glycogen content in cysts)

* Pneumocystis carnii is non cultured on SDA

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PrepLadder

OCCUPATIONAL DISEASES

02:32:29

FARMER'S LUNG

- d/t Hay dust (stored Hay)
  - Caused by Micro / Polyspora faenii, Faenia rectivirgula, Thermo actinomyces vulgaris
- Type III hypersensitivity

BAGGASSOSIS

- d/t Sugar Cane Dust
- Caused By Thermo Actinomyces Sacchari

MALTSTER'S LUNG

- d/t Barley & Malt
- Caused by Aspergillus clavatus

CHEESE WASHER'S LUNG

- a/w Cheese factory workers
- Caused by Penicillium caseiocolum

MAPLE BARK STRIPPER'S LUNG

- d/t Maple tree bark
- Caused by Cryptostroma corticale

SEQUIOSIS

- d/t Redwood saw dust
- Caused by Aureobasidium pullulans

SUBEROSIS

- d/t Cork
- Caused by Penicillium frequentans

WOOD PULP WORKERS

- d/t Wood pulp
- Caused by Alterneria species

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HUMIDIFIER'S LUNG

- d/t Humidifier
- Caused by Thermoactinomyces vulgaris , Thermoactinomyces candidus

Mycotoxins

02:37:21

- Used in Bioterrorism
- Iraq used Aflatoxin as warfare agent
- Soviet Union used Trichothecenes (caused yellow rain)

1. Aflatoxins – secreted by Aspergillus flavus
   - Leads to HCC (Hepatocellular Ca)
2. Citrinin
3. Ergot alkaloids
   - Secreted by Claviceps purpurea
   - Edibles + Claviceps purpurea seeds → Ergotoxicosis
   - Consumption of contaminated edibles leads to
     i) St. Vitus Dance &
     ii) St. Anthony's fire
   - Gastric irritation occurs

     ↓

     Restlessness [St. vitus Dance]
4. Fuminosins
5. Ochratoxin
6. Patulin
7. Trichothecenes
8. Zearalenone

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Subcutaneous Mycosis

Mycetoma

Clinical features :

Triad of :

1) Swelling
2) Discharging sinuses
3) Granules in discharge
   - Affect lower and upper extremities

Actinomycetoma

Gram positive Branching filamentous bacteria

- Nocardia, Actinomadura
- Numerous sinus & Swelling
- Purulent discharge
- Tt: Cotrimoxazole

Eumycetoma

- Fungal
- Madurella species
- Single sinus & Swelling
- Serous discharge

Tt → Itraconazole
→ Amphotericin B

Sporotrichosis

- Caused by Sporothrix schenckii
- Thorn prick (Gardener's disease)
- Dimorphic fungus
- Mold shows flower like sporulation
- Cigar shaped yeast cells
- Asteroid bodies

Chromoblastomycosis

- Thorn prick/ wood prick
- Verrucous lesion/ warty lesion
- Biopsy : Sclerotic bodies/ medlar bodies
  - Copper penny appearance

Rhinosporidiosis

- Caused by Rhinosporidium seeberi
- Bilateral nasal polyps

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Unit: 4 Parasitology

Chapter 9

PROTOZOOLOGY

PROTOZOA

00:00:39

Classification

AMOEBAE	CILATES
a Free living – Naegleria Fowleri – Acanthmoeba – Balamuthia b Intestinal – Entamoeba histolytica – Entamoeba coli	Balantidium coli
FLAGELLATES	**SPOROZOA**
a Intestinal – Giardia lamblia b Vaginal – Trichomonas vaginalis c Blood & Tissue – Leishmania – Trypanosoma	a Blood species – Plasmodium – Babesia b Tissue species – Toxoplasma c Intestinal species – Isospora – Cyclospora

Entamoeba hystolytica

00:07:20

QUADRI NUCLEATE CYST [Infective Form]

↓

Ingested with contaminated food & water

↓

Bypasses gastric acidity

↓

E. histolytica Quadrinuclea Cyst

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Excystation in small intestine / large intestine

↓

TROPHOZOITES

- Active feeding & growing stage
- 8 Metacystic Trophozoites are formed

Favorable Conditions

1. 85% → ASYMPTOMATIC
2. 5-15% → INTESTINAL AMOEBIASIS
3. EIA (Extra Intestinal Amoebiasis)

Unfavorable Conditions

↓ Encystation

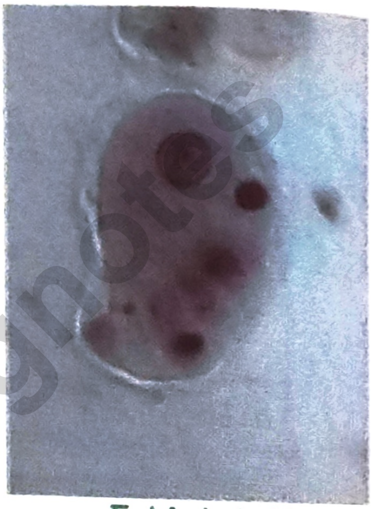
Uninucleate cyst

↓ Binary fission

Binucleate cyst

↓ Binary fission

Quadrinucleate cyst (faeces)



E. histolytica

EIA [EXTRA INESTINAL AMOEBIASIS]

↓

Trophozoites secretes Cysteine proteinase

↓

Erode intestinal layers

↓

Flask Shaped Ulcers

↓ Via portal circulation

Liver [Extra Intestinal Amoebiasis]

- MC site For EIA → Liver > Lungs > Brain
- Chocolate pus/ Ancovy sauce pus formed due to damage to Hepatocytes
- Right lobe (Posterio superior portion) is involved
- IOC For EIA → ELISA

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PrepLadder

00:17:45

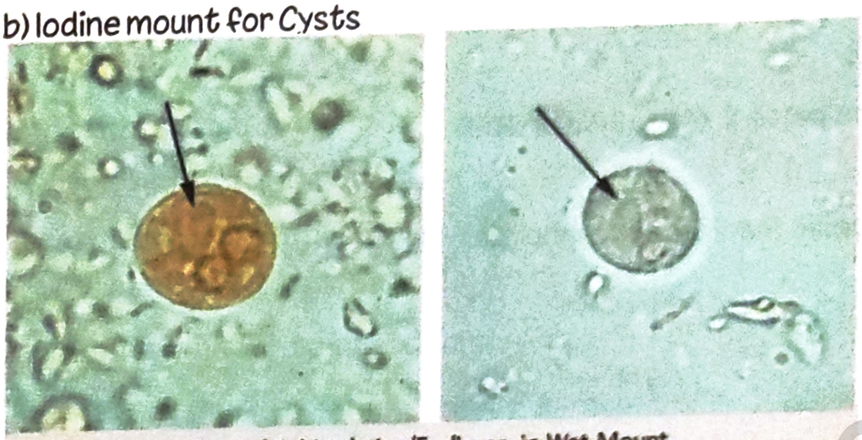
INTESTINAL AMOEBIASIS

LAB DIAGNOSIS

1. Stool Examination

   a) Saline mount for Trophozoite

   b) Iodine mount for Cysts



Cyst of E. histolytica/E. dispar in Wet Mount.
Left Side: Stained with Iodine
Right Side: Unstained preparation (Arrow- Notice the chromatoid body with blunt, rounded ends)

(a) Stool sample + Normal saline→ Trophozoites seen under microscope

- Motile by Pseudopodia
- Eccentric Nucleus with central Karysome is present
- ERYTHROPHAGOCYTOSIS seen
  - Engulfed RBCs
  - Seen in fresh stool samples
  - Characteristic features of E. hystolitica

(b) Stool sample + Iodine mount → cyst is seen under microscope

- 4 nuclei cyst (mostly)
- Uninucleate or binucleate cysts with chromatoidal bars & Glycogen vacuoles are also seen sometime

2. CULTURE

Media used are

P → PHILIPS MEDIA

C → CRAIGE'S MEDIA

R → ROBINSON MEDIA

3. PCR For Entamoeba hystolytica

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PrepLadder

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Or click this page to join

NAEGLARIA FOWLERI [Brain Eating Amoeba] 00:26:37

- Infective form → Amoeboid form ⟷ Trophozoite Form

Amoeboid Form > Trophozoite form

↓

Enters Nares

↓

Erodes Cribriform plate & enter into brain

↓

PAM [Primary Amoebic Meninogoencephalitis]

- H/o swimming in Pond +nt

LAB DIAGNOSIS

CSF EXAMINATION

- Trophozoites of N. fowleri are seen
- Neutrophils present

ACANTHAMOEBA SPECIES 00:30:17

- Causes CONTACT LENS KERATITIS
- Aerosol route

↓

Lungs

↓ Hematogenous spread

Brain

↓

Granulomatous Encephalitis

*Spread can occur even through Optic route

LAB DIAGNOSIS

CSF Examination

→ Cysts & Trophozoites are seen

→ lymphocytes present

ACANTHAMOEBA

FREE LIVING AMOEBAE	
Naeglaria fowleri Acanthamoeba Balamuthia	} causes Encephalitis

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PrepLadder

INTESTINAL FLAGELLATES

GIARDIA LAMBLIA

00:33:13

- Quadrinucleate cyst [Infective Form]

  ↓

  Ingested with contaminated food & water

  ↓

  Bypass gastric acidity

  ↓

  Excystation mostly in duodenum

  ↓

  Trophozoites attach to intestinal surfaces

  ↓

  Malabsorption Syndrome

  Steatorrhoea (Fat in stools, Bulky stools)

TROPHOZOITE

- TENNIS RAQUET APPEARANCE OR
  MONKEY FACE APPEARANCE OR
  TEAR DROP APPEARANCE

Giardia Trophozoite in Stool

- Axostyles are present
  4 pairs of Flagella → FALLING LEAF LIKE MOTILITY
  Sucking disc → helps in adherence to various surfaces (Virulence factor)

CYST

- 4 nuclei +nt
- have Axostyle
- ENTERO TEST/ STRING TEST done to demonstrate Trophozoites
- Giardia lamblia infection a/w CVID [Common Variable Immuno Deficiency]

Giardia Cyst

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VAGINAL FLAGELLATES

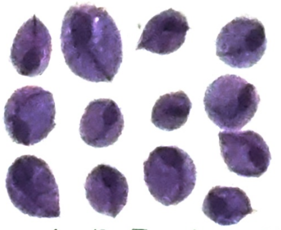
TRICHOMONAS VAGINALIS

00:42:47

- No cystic stage
- only Trophozoite stage is present
  - Trophozoite have 5 pairs of Flagella → Twitching Motility
- Causes TRICHOMONIASIS

FEATURES

- pH > 4.5
- Greenish frothy discharge
- Colpitis macularis / Strawberry vagina



T.vaginalis Trophozoites

LAB DIAGNOSIS

1. URINE EXAMINATION → Motile Trophozoites are found
2. CULTURE → on BUSHLEY'S, JOHNSON & TRUSSEL MEDIA
   - DOC → METRONIDAZOLE to both partners

HAEMO FLAGELLATES

1. LEISHMANIA → Promastigote [Infective Form]
2. TRY PANOSOMA → Metacylic Trypomastigote [Infective Form]

LEISHMANIA

00:47:44

- L. donovani → causes Kala Azar [Vector → Phlebotomous (Sand Fly)]
- L. Tropica → causes Oriental sore/ Delhi boil or Baghdad boil/ Aleppo button
- L. brasilensis → causes Espundia [Muco cutaneous leishmaniasis]

Sand Fly

KALA AZAR

CLINICAL FEATURES

- Hyper pigmentation + Fever + Hepatosplenomegaly seen
- KA with HIV +ve → Hepatosplenomegaly absent (atypical presentation)
- Anemia, Thrombocytopenia, Leucopenia seen
- Hypergamma globulinemia seen

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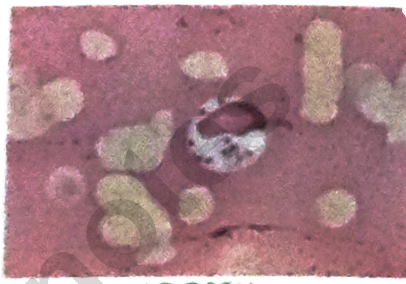
LAB DIAGNOSIS

1. BLOOD SMEAR

- A → Anemia
  L → Leucopenia
  T → Thrombocytopenia

Kala Azar in children


LD BODY

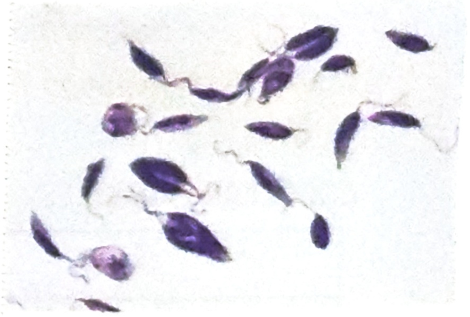
- Hypergammaglobunemia demonstrated by
- NAPIER'S ALDEHYDE TEST
- CHOPRA'S ANTIMONY TEST

2. SPLENIC BIOPSY VS BONE MARROW BIOPSY

- Splenic Biopsy is more sensitive
- More Amastigote Forms are observed
- Aflagellar Forms aka LD BODIES [LEISHMAN DONOVAN BODIES] are seen

NNN MEDIA

3. rk - 39 ANTIGEN TESTING using card Method
4. MONTENEGRO TEST [Skin Testing]
5. CULTURE ON NNN [Novy Neil Nicholle] media
   - Promastigote Forms (Flagellar Forms) demonstrated
6. SEROLOGICAL TESTS are available
7. PCR For L. donovani is available
8. DOC → LIPOSOMAL AMPHOTERICIN B


PROMASTIGOTES

PKDL [POST KALA AZAR DERMAL LEISHMANIASIS]

↓

After 2 yrs of successful Rx

↓

Patient suddenly develops crops of nodules / macules over the skin (d/t Dermotropic conversion)

PKDL

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PrepLadder

TRYPANOSOMA BRUCEI

00:59:26

- Gambiens
  - Causes West African Sleeping sickness
  - Winter bottom sign seen (B/L Cervical lymphadenopathy)

- Rhodesiense → East African Sleeping sickness
- TSE TSE FLY [Glossina Species] transmits sleeping sickness
- AFRICIAN TRYPANOSOMIASIS = Sleeping sickness

TSETSE FLY

TSETSE FLY

T. CRUZI

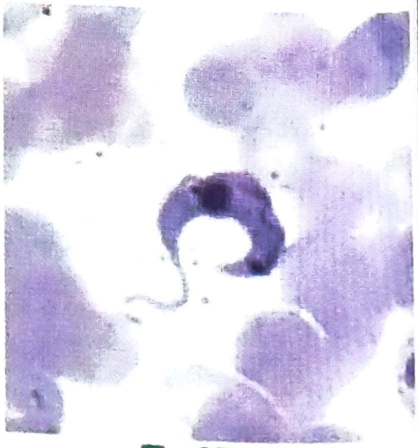
01:02:06

REDUVID BUG

- Transmitted by Reduvid bug
- Causes CHAGAS DISEASE/AMERICAN TRYPANOSOMIASIS

FEATURES

- CHAGOMA → Entry site Edema
- ROMANA SIGN → Eye lid swelling
- M → Mega oesophagus
- M → Mega colon
- M → Myocarditis
- M → Meningo encephalitis

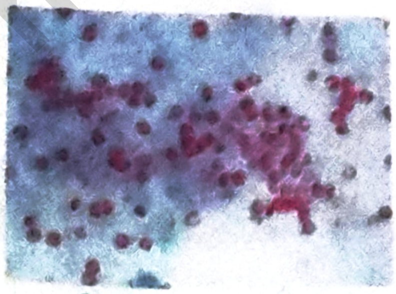


T. CRUZI

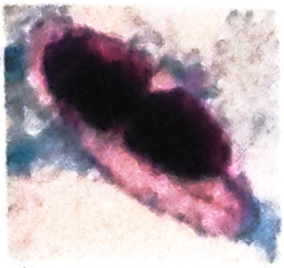
COCCIDIAN PARASITES

01:05:58

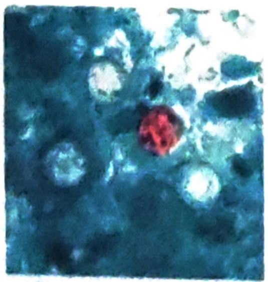
	Oocyst Size
Cryptosporidium parvum	4-6 μ
Cyclospora	8-10 μ
Isospora belli	23-36 μ



Cryptosporidium



Isospora belli



Cyclospora

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- All are Acid fast
- if Stool Examination shows Coccidian parasites

↓

Check HIV status

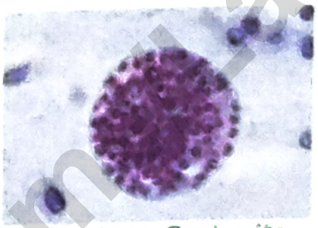
- +ve → Admit
  - IV Fluids
  - Antibiotics
- -ve → self limiting diarrhoea

TOXOPLASMA GONDII

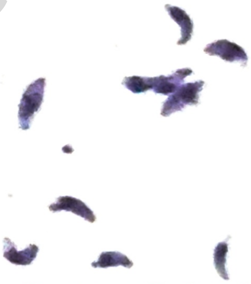
01:09:56

- Causes Toxoplasmosis
- Definite Host → Cat
- Intermediate Host → Rat/Man
- Cat → Enteric cyclic occurs → Oocyst formed
- Rat/Man → Exoenteric cyclic occurs → No Oocyst formation

- Infective forms
1. Sporulated oocyst
2. Tachyzoites
3. Bradyzoites



Toxoplasma Bradyzoite



Toxolasma Tachyzoites

*Oocyst is not the infective form of T. gondii

*Oocyst having Sporozoites (Sporulated Oocyst) is infective form

- LAB DIAGNOSIS

1. Blood Smear + Geimsa Stain	Tissue Smear + Geimsa Stain
Tachyzoites - Rapidly dividing forms - Sickle shaped - Seen in blood	Bradyzoites - Slowly dividing forms - Seen in Tissue cysts

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PrepLadder

2. SABIN-FELDMAN DYE TEST → Compliment fixation test
3. PCR For Toxoplasma gondii
4. ELISA in a pregnant woman with Toxoplasmosis

- if she have IgM antibodies (Acute infection)
  - Cause for concern
- if she have IgG antibodies (Chronic infection/ Vaccination)
  - No cause for concern
- Toxoplasmosis in immunocompetent → mild Cervical lymphadenopathy
- Toxoplasmosis in immunodeficiency (HIV +ve) → Seizures
- Toxoplasmosis in pregnancy

- at 1st Trimester → max damage to fetus
- at 3rd Trimester → max chance of transmision

PLASMODIUM SPECIES

01:20:39

- P. vivax
- P. ovale
- P. falciparum
- P. malariae
- P. knowlesi (In Indonesia & Malaysia)

Anopheles

LIFE CYCLE

SPOROZOITES

↓

Bite of female Anopheles mosquito

↓

Sporozoites move towards liver

D/t presence of Circum Sporozoite Proteins (CSP) in liver

↓

Pre Erythrocytic Schizogony

↓

Merozoites

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Merozoites

- Some change to

HYPNOZOITES

- Resting Forms
- Reside in liver
- Responsible for Relapse
- Seen in P.vivax,
- DOC → PRIMAQUINE

Some capture RBC

↓

Erythrocytic Schizogony

↓

Trophozoites

↓

Schizont

↓

Merozoites

↓

Gametocytes

(Micro/Macro)

↓

Female Anopheles bites man

& take Blood meal

↓

Mature gametes

Female + Male

↓

Zygote

↓

Ookinete

- Needle like structure
- penetrate gut of mosquito

↓

Oocyst

↓

Moves to Salivary glands

↓

Sporozoites

↓

Proboscis of mosquito

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PrepLadder

Conditions preventing P. falciparum infection

- G → G_6PD Deficiency
- S → Sickle cell anemia
- T → Thalassemic trait
- Excess of Fetal Hb

Condition preventing P.vivax infection → Duffy negative RBC

LAB DIAGNOSIS

1. Blood smear Examination
   - a) Thick smear → to know Prevalence of parasite
   - b) Thin smear → to know Speciation

P. vivax	P. falciparum
Young RBCs affected	All age RBCs affected
RBC Size ↑ ses	N Size RBCs
SCHUFFNER'S DOTS SEEN	MAURER'S DOTS seen
ACCOLE FORMS –nt	ACCOLE FORMS +nt
SCHIZONT STAGE +nt	SCHIZONT STAGE –nt
Gametocyte → No Proper Shape	Gametocyte → Banana shaped
Relapse seen d/t Hypnozoites	Recrudescence seen d/t incomplete Rx
Single large ring within RBC	Multiple rings are seen within RBC

P.malariae

Z → Ziemann's dot

O → Old age RBC

O

M → Max. incubation period

I

N → Nephrotic syndrome

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Prep

2. BUFFY COAT EXAMINATION

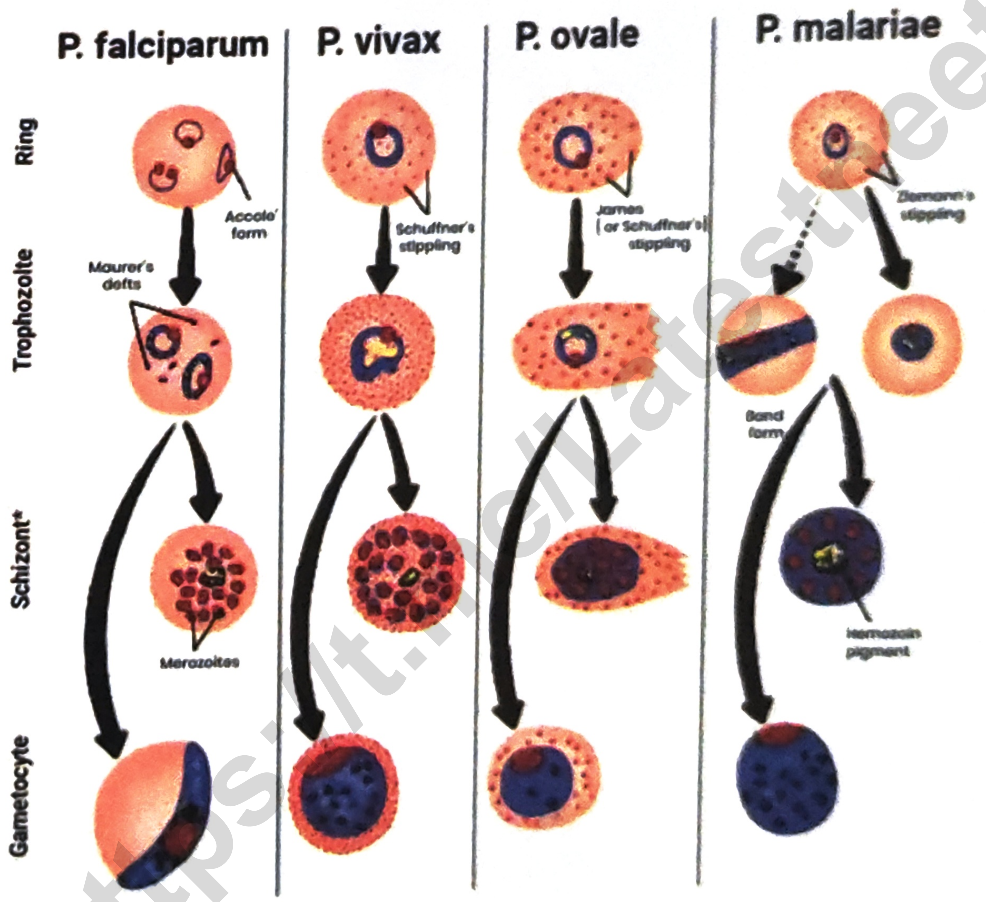
- using Acridine orange & using Fluorescent Microscopy

3. CULTURE on RPMI-1640 media

4. MALARIA CARD TEST

- HRP-2 based kits [Histidine Rich Protein - HRP] for P. falciparum
- LDH & Aldolase based Kits For other plasmodium species

5. PCR For Malarial Parasites



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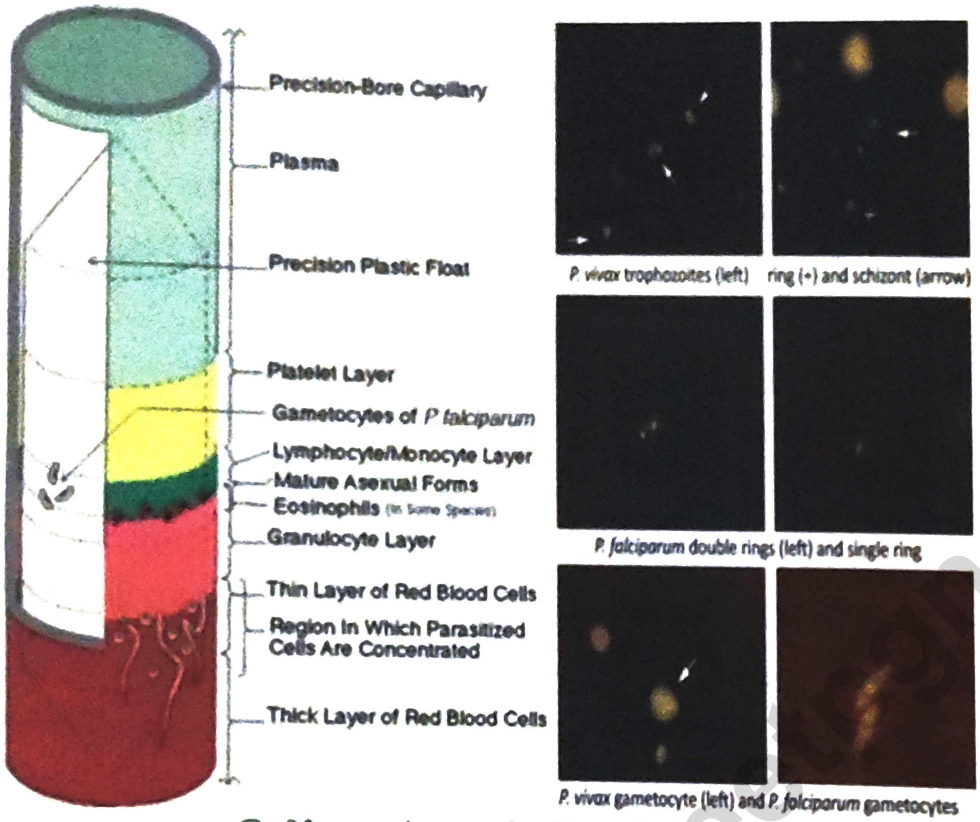
PrepLadder



Buffy coat examination of plasmodium

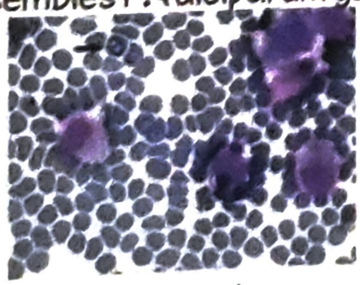
Malaria Card Test

01:55:01

BABESIA MICROTI

- Intra erythrocytic protozoa
- Causes BABESIOSIS
- Transmitted by TICK
- MALTESE CROSS ARRANGEMENT in RBC
- Resembles P. falciparum gametocyte

Tick



Babesia

Maltese Cross Arrangement

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BALANTIDIUM COLI

01:57:39

- Ciliate
- Contains
  - Peristome
  - Cytostome
  - Cytopharynx
  - Micro & Macro Nucleus
- Largest intestinal protozoa
- Rotatory motility +nt
- Causes Diarrhoeal Episodes
- DOC → TETRACYCLIN

Balantidium coli Trophozoite

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PrepLadder

Chapter 10

HELMINTHOLOGY

CESTODES	TREMATODES	NEMATODES
→ Tape like → Monoecious	→ Leaf like → Monoecious except Schistosomes (Dioecious)	→ Cylinderical → Dioecious
→ Suckers with hooks	→ Suckers without hooks	→ Have Buccal capsule
→ Absent alimentary canal	→ Poorly developed alimentary canal	→ Well developed alimentary canal
→ Body cavity absent	→ Body cavity absent	→ Body cavity present

CESTODES

00:04:15

BASIC STRUCTURE

SCOLEX	NECK	STROBILA
Consists of Suckers (Rostellum & Hooklets)		Contains Proglottids

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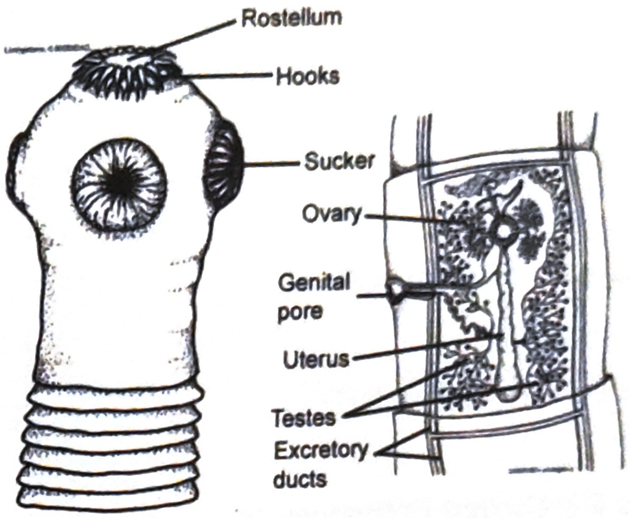

CLINICALLY IMPORTANT CESTODES

	Definite Host	Intermediate Host
Taenia solium	Man	Pig
Taenia saginata	Man	Cattle
Echinococcus granulosus	Dog	Sheep / Man
H. nana	Man	Man
Diphyllobothrium latum	Man	1st – Cyclops 2nd - Fish

DIPHYLLOBOTHRIUM LATUM/ FISH TAPE WORM 00:08:10

Plerocercoid larva [L_3 larvae] [Infective Form] present in Fish

↓

Man eats infected Fish

↓

Larvae released

↓

Changes to Adult Forms

↓

Settle up in small & large intestine

↓

↓ Vit. B_{12} absorption → Megaloblastic Anemia

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PrepLadder

↓

Adult Forms

↓

Produce eggs

↓

Goes into water

↓

Hatch into Coracidium larvae [L_1 Larvae]

↓

Coracidium larvae reaches Cyclops & in Cyclops it changes to Procercoid [L_2 larvae]

↓

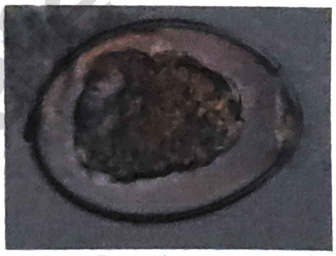
Eaten by Fish

↓

In Fish it changes into Plerocercoid larvae [L_3 larvae]

*Egg of Diphyllobothrium latum → Operculated egg

Cyclop



D. latum egg

H. nana / Dwarf Tape Worm 00:14:55

Eggs are ingested with contaminated food & water [Infective form]

↓

Eggs hatch in intestine

↓

Cysticercoid larvae released

↓

Attach to the intestinal layers

↓

Changes to Adult form

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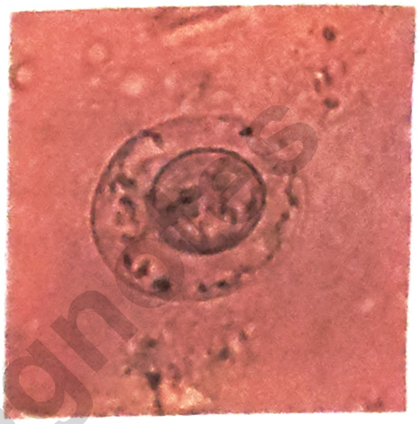
PrepLadder

↓

Produce eggs

Eggs passed out In Faeces	Eggs hatch within lume
	↓
	Releases Hexacanth embryo
	↓
	Penetrates back into circulation
	↓
	Auto infection



EGG

- Contains
  - Polar filaments
  - Central hooks
- Non bile stained Egg

Non Bile Stained Eggs
N → Necator americanus E → Enterobius vermicularis H → H. nana A → Ancylostoma duodenale

Rx → DOC → PRAZIQUANTEL

ECHINOCOCCUS GRANULOSUS/ DOG TAPEWORM 00:20:48

Eggs of E. granulosus ingested with contaminated Food & water [Infective form]

↓

Eggs hatch in duodenum

↓

Releases Hexacanth embryo

↓

Penetrates the intestinal layer

↓

Through Portal circulation

↓

Reaches Liver

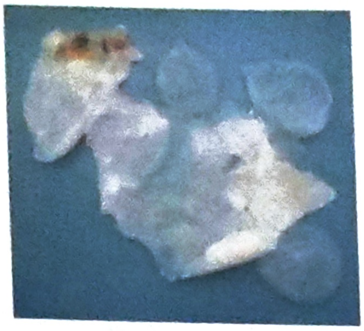
↓

Forms Hydatid cyst

HYDRATED CYST

00:24:05

- Consists of
  - Pericyst
  - Ectocyst
  - Endocyst (Germinal epithelium)
    - Most active layer
    - Secretes Hydatid fluid
    - a/w Brood capsule, Protoscolices



Hydatid cyst

Hydatid Fluid

- Casoni's Test
  - Type 1 hypersensitivity
    - Immediate Wheal & Flare reaction
- Obsolete now
- E.multilocularis → causes Malignant Hydatid Disease

TREATMENT OF HYDATID CYST

1) Mild to Moderate Size Cyst

PAIR THERAPY → Per cutaneous Aspiration Injection Reaspiration therapy Using Scolicidal agents

2) Large Size cyst → Surgical Removal

Taenia solium	Taenia saginata
→ Shorter	→ Longer [in metres]
→ Armed tapeworm (Rostellum & hooklets present)	→ Unarmed tapeworm (No Rostellum & hooklets)
→ Eggs of both look like same DD-T. Saginata eggs hooklets are acid fast	

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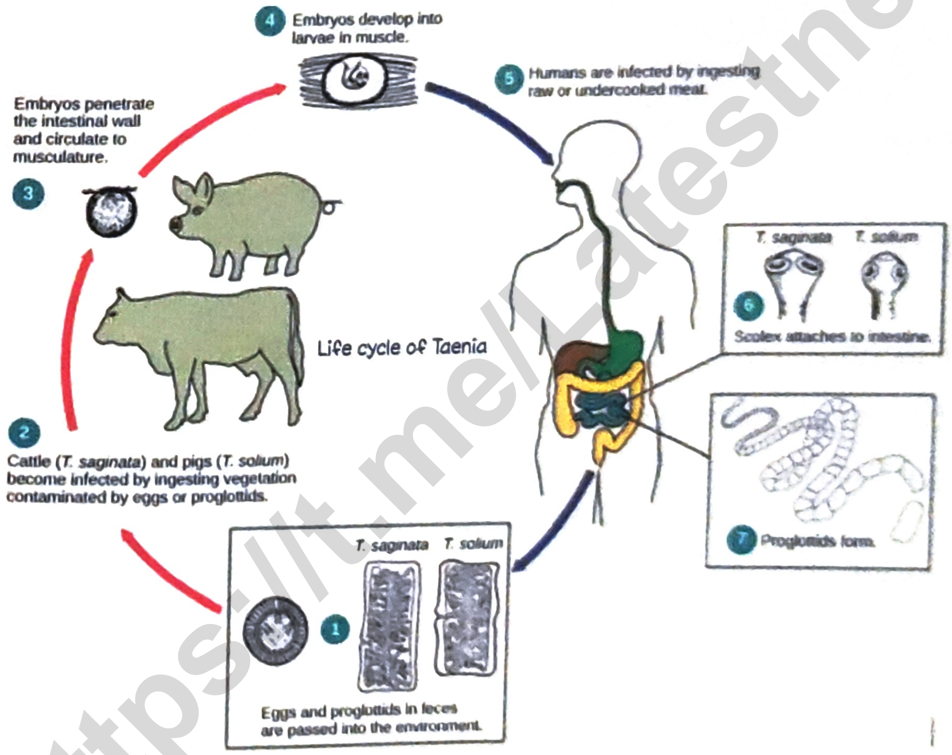
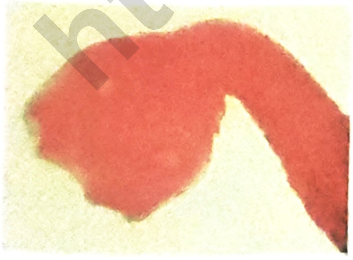
PrepLadder

Taenia solium	Taenia saginata
→ Proglottids number → <1000 Proglottids shed off	→ Proglottids number → >1000 Proglottids shed off
→ Eaten uncooked pork with Cysticercus cellulosae ↓ – TAENIASIS [Diarrhoeal episodes] – CYSTICERCOSIS [Few cases] (d/t ingestion of eggs)	→ Eaten uncooked beef with Cysticercus bovis ↓ – TAENIASIS [Diarrhoeal episodes] – Nothing happen & if eggs are eaten

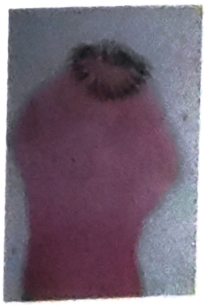
* Proglottids → Structure in which Male & Female ill defined organs are present



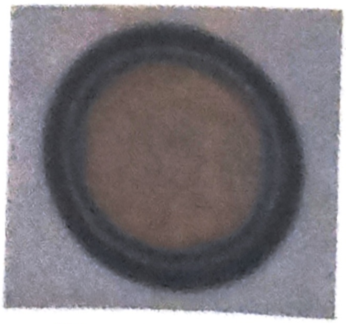
Life cycle of Taenia



T. saginata Scolex



T. solium scolex



Taenia egg

PrepLadder

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CYSTICERCOSIS

Portal of entry for Cysticercosis

↓

Eggs with contaminated food & water

↓ can lead to

NEUROCYSTICERCOSIS

→ MC site → Sub arachnoid > Parenchymal

→ Presents with Atypical Seizures

→ IOC → CT/MRI Scan

→ DOC → ALBENDAZOLE

TREMATODES/ FLUKES

00:41:50

Properties

1) Leaf like
2) Monoecious
3) Have two Intermediate hosts
   - 1st → Snail
   - 2nd → Aquatic Vegetation
     - Fish
     - Crab/Cray Water Fish
4) Infective Form → Meta cercariae (cyst like structure)
5) All have Operculated Eggs [Lid]

EXCEPTION → SCHISTOSOMES

- Dioecious
- Single intermediate host → Snail
- Infective form → Cercariae
- Have non operculated eggs

SCHISTOSOMES

00:46:23

SCHISTOSOMA HAEMATOBIUM

- resides in Vesical plexus
- causes SCC of Urinary bladder > Transitional cell Ca
- Causes Terminal Hematuria

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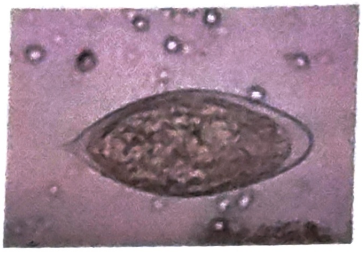
PrepLadder

- Egg has Terminal spine
- Eggs are excreted in urine

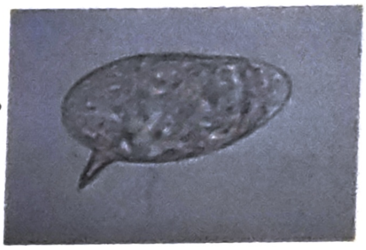


Schistosoma hematobium

* SCC → Squamous cell carcinoma

S.MANSONI

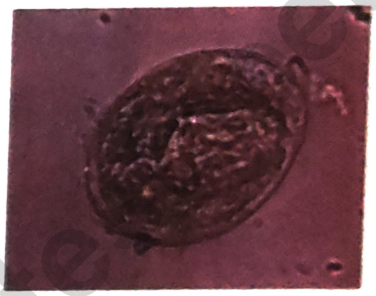
- resides in Inferior Mesenteric plexus
- causes Swimmer's itch & Katayama fever
- Eggs are excreted in faeces



Schistosoma mansoni

S. japonicum

- resides in Superior Mesenteric plexus
- causes Katayama Fever & Eosinophillic diarrhea
- Eggs are excreted in faeces



Schistostoma japonicum

Fasciola Hepatica / Sheep liver Fluke

00:52:35

- 1st Intermediate Host → Snail
- 2nd Intermediate Host → Aquatic vegetation [with Metacercariae]

↓

Man eats this aquatic vegetation

↓

Metacercaria is ingested

↓

In duodenum excystation occurs

↓

Larvae released

↓

Penetrates intestinal layers

↓

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PrepLadder

Penetrates Liver capsule

↓

Penetrates bile ducts

↓

Matures to Adult Forms

↓

Produce Eggs

↓

Eggs passed out in faeces

↓

Comes in contact with water

↓

Eggs → Miracidum [In SNAIL]

↓

Sporocyst

↓

Radiae [1st generation & 2nd generation]

↓

Cercariae

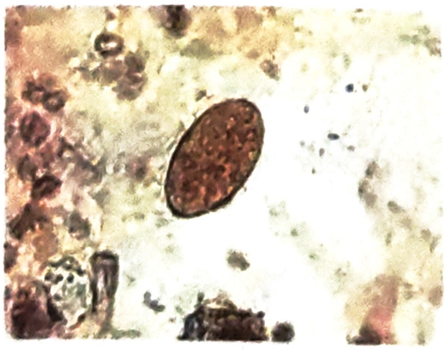
↓

Metacercariae [cyst like structures]

↓

Goes & settles on aquatic vegetation

Fasciola hepatica



F. hepatica egg (Operculated)

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PrepLadder

Clonorchis sinensis/ Oriental liver fluke / Chinese liver fluke 00:59:18

1st IH → Snail

2nd IH → Fish [with Metacercariae]

↓

Eaten by man

↓

Excystation

↓

Larvae released

↓

Penetrates intestinal layers

↓

Penetrates Liver capsule

↓

Penetrates bile ducts

↓

Matures to adult forms & causes constant inflammation & fibrosis of bile ducts

↓

Causes Cholangiocarcinoma [Bile duct carcinoma]

Paragonimus westermanii [Lung Fluke] 01:03:23

1st IH → Snail

2nd IH → Crab/Cray water Fish with Metacercariae

↓

Eaten by man

↓

Metacercariae ingested

↓

Excystation in Duodenum

↓

Larvae released

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PrepLadder

↓

Penetrates intestinal layers

↓

Penetrates diaphragm

↓

Penetrates Rt. Lung capsule

↓

Enters into Rt. Lung (mostly)

↓

Cysts → Erosions

↓

Red Brown sputum

(Endemic Hemoptysis)

NEMATODES

01:06:53

SMALL INTESTINAL NEMATODES	TISSUE NEMATODES
Ascaris Hook worm Necator americanus Strongyloides Stercolaris	Resides in Lymphatics Wuchereria bancrofti Brugia Malayi Brugia timori
LARGE INTESTINAL NEMATODES Trichuris trichiura Enterobius vermicularis	Resides in Subcutaneous sites Loa loa Onchocerca volvulus
MISCELLANEOUS NEMATODES Dracunculus medinensis Trichinella Spiralis	Resides in Serous cauities Mansonella Ozzardi Mansonella perstans

TRANSMISSION BY

EATING EGGS	PENETRATION OF SOLE OF FOOT (Filariform larvae)
E - Enterobius vermicularis A - Ascaris T - Trichuris Trichiura	A - Ancylostoma duodenale N - Necator americanus S - Strongyloides Stercolaris

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PrepLadder

LUNGS FOR MATURATION

A² → Ancylostoma duodenale
Ascaris
N → Necator americanus
S → Strongyloides stercolaris

Large Intestinal Nematodes

Trichuris trichiura / WHIP WORM

01:15:40

Eggs are ingested with contaminated Food & water

↓

Hatch up in duodenum or caecum

↓

Larvae released

↙ ↘

Male worms   Female worms

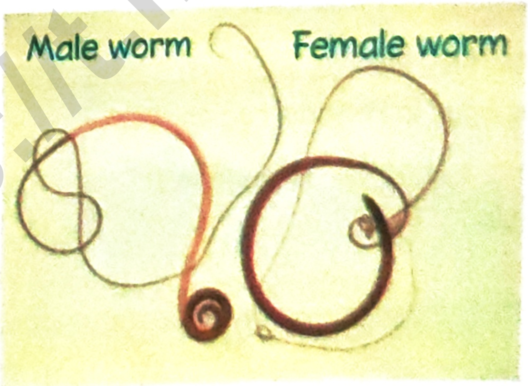
⊕ Fertilise

↓

Eggs

↓

Passed out in Faeces



EGG

- Barrel/dumb bell shaped egg
- have mucous plugs on both sides
- causes Fe deficiency anemia

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PrepLadder

- causes Frequent Rectal prolapse in children

↓

Coconut Cake Like Rectum

↓

Growth retardation

Egg of Trichuris

ENTEROBIUS VERMICULARIS / PIN WORM / THREAD WORM / SEAT WORM / OXYURIASIS

01:20:52

- Presents with Perianal pruritis

Eggs deposit in nail beds

↓

Child putting up fingers in mouth

↓

Eggs are ingested

↓

Hatch up in Small Intestine/ Large Intestine

↓

Larvae released

Male worms   Female worms

+ Fertilise

↓

After fertilization, Male dies

Female goes in night time & lays eggs in Perianal region

↓

Autoinfection

AUTO INFECTION SEEN IN

C → Capillaria phillipensis

→ Cryptosporidium parvum

H → H. nana

E → E. vermicularis

S → Strongyloides stercolaris

T → Taenia solium

E. vermicularis

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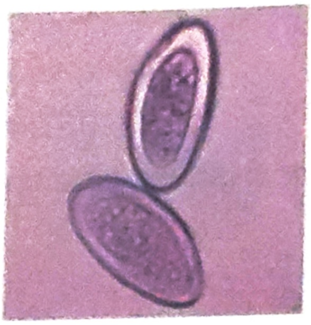
PrepLadder

EGG

- Plano convex egg
- having a Tadpole like larvae
- non bile stained egg



Enterobius egg

NIH SWAB

- National Institute Health Swab
- Made up of glass rod with cellophane tape

NIH swab

SMALL INTESTINAL NEMATODES

ANCYLOSTOMA DUODENALE / HOOK WORM 01:30:28

Penetration of Filariform larvae in sole of foot

↓

Causes Ground itch/ Maculopapular dermatitis

↓

Penetrates all skin layers

↓

Gets into Cutaneous circulation

↓

Lymphatics

↓

Venous circulation

↓

Portal circulation

↓

Liver

↓

Rt. Side of heart

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PrepLadder

↓

Lungs

↓

After slight maturation

↓

RETROGRADE JOURNEY

↓

Penetrates Pulmonary capillaries

↓

Alveoli → Bronchiole

↓

Larynx ← Trachea

↓

Pharynx

↓

Swallowed with sputum

↓

In the Esophagus, 3rd moulting happens

↓

In duodenum, 4th moulting happens

↓

Mature larvae

Male worms   Female worms

⊕Fertilise

↓

Eggs

↓

Eggs passed out in Faeces

↓

Soil → Eggs

↓

Rhabditiform larvae

[2 moultings (1st & 2nd)]

Rhabditiform Larva

Ankylostoma egg

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PrepLadder

↓

Filariform larvae [Needle like]

[Infective Form]

A. Brasilensis

Animal worm

↓

Can not penetrate all skin layers

↓

Trapped b/w skin layers

↓

Cutaneous larvae migrans

Cutaneous larvae migrans

A. Duodenale

- causes Fe deficiency anemia [0.2 ml/Day/worm Blood Sucked]
- EGG
  - Segmentation is seen [Blastomeres]
  - CHANDLER'S INDEX→ 300 eggs/gm of Stool
    - Social problem

S. Stercolaris

01:42:30

- Strongly a/w HIV +ve
- OVO - vivi parous
- Undergo Auto infection

↓

Causes HYPER INFECTION SYNDROME in HIV +ve

- CNS & viscera involved
- Carries some mortality

Strongyloides stercoralis

- Undergoes PARTHENOGENETIC DEVELOPMENT
  - Females directly lay eggs without male

- also undergoes Cutaneous larvae migrans → LARVAE CURRENS
  - Moves faster under skin

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PrepLadder

ASCARIS

01:46:20

Eggs are ingested with contaminated food & water

↓

Hatch up in duodenum

↓

Larvae released

↓

Penetrates intestinal layers

↓

Gets into Portal circulation

↓

Liver

↓

Rt. Side of heart

↓

Lungs [2nd & 3rd moulting happens

↓

RETROGRADE JOURNEY

↓ (penetrating pulmonary capillaries)

Alveoli → Bronchiole

↓

Larynx ← Trachea

↓

Pharynx

↓

Swallowed with sputum

↓

In duodenum, 4th moulting occurs

↓

Mature larvae

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PrepLadder

Mature larvae

Male worms — Female worms

⊕Fertilise

↓

Eggs

↓

Passed out in Faeces → Soil

↓

Eggs [1st moulting]

↓

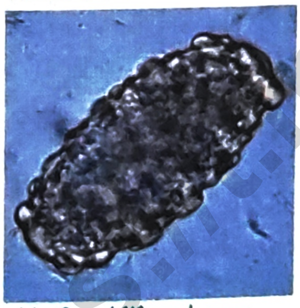
Rhabditiform larvae

↓

Eaten by man

- Ascaris causes Intussusception → Intestinal obstruction
- Strongly a/w Loeffler's syndrome → Massive Eosinophilia

EGG OF ASCARIS



Unfertilized egg

Fertilized egg

EGGS WHICH FLOAT ON SATURATED SALT SOLUCTION

F → Fertilised egg of Ascaris

A → Ancylostoma duodenale

T → Trichuris trichiura

E → Enterobius vermicularis

H → H. nana

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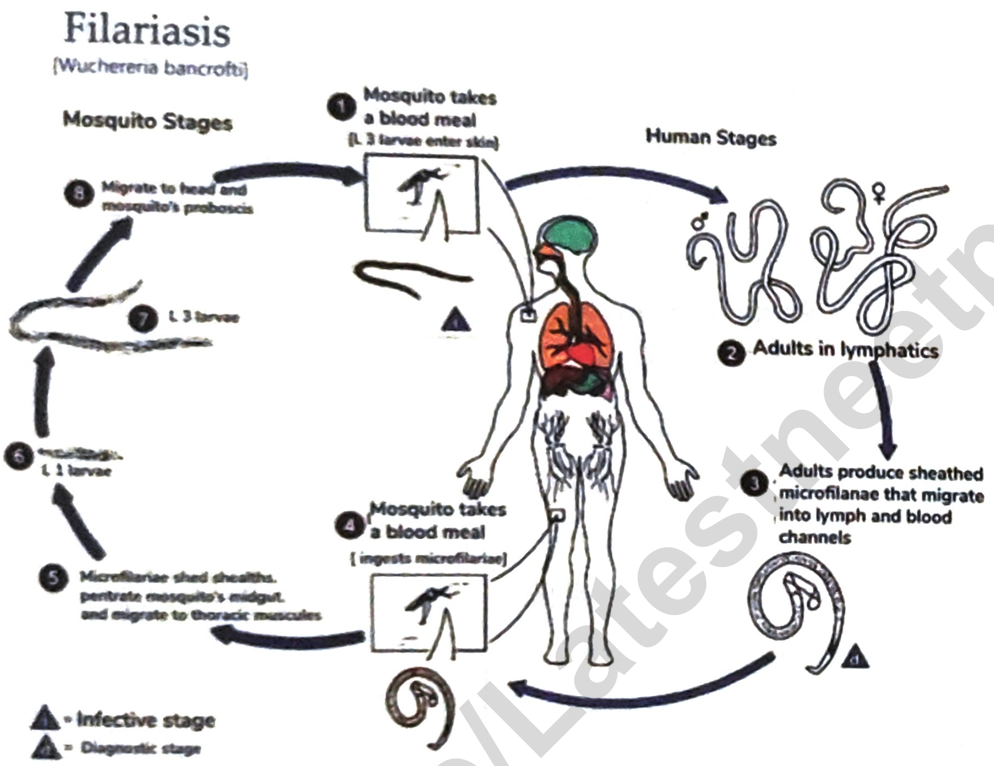
TISSUE NEMATODES

WUCHERERIA BANCROFTI

01:56:30

- INFECTIVE Form → L_3 Filariform larvae
- Transmitted by
  - Culex [mainly]
  - Aedes
  - Anopheles



Acute Filariasis Features → Fever, lymphadenitis, lymphangitis

Chronic Filariasis Features

- Elephantiasis
- Hydrocele
- Granuloma breast
- Kidney LN block up → Chyluria, Albuminuria

LAB DIAGNOSIS

1) BLOOD SMEAR EXAMINATION
   - Taken at night time d/t Nocturnal periodicity

2) DEC PROVOCATIVE TEST
   - DEC irritates Wuchereria & Wuchereria comes in blood

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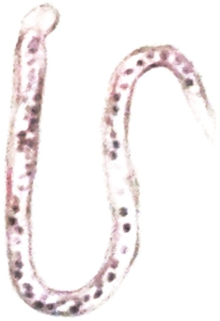
↓

Do blood smear examination

↓

Look for Microfilariae (Tail Tip)



Wuchereria bancrofti

Elephentiasis

TAIL TIP

- Terminal Tail Tip is examined for identification
- No Nuclei seen in W. bancrofti
- Tail Tip with 2 terminal Nuclei → Microfilariae of Brugia malayi

Brugia malayi

3) RADIOLOGICAL

- X ray
- CT Scan
- USG of Scrotum → DANCING WORM SIGN Seen

Hydrocele

4) PCR For W. bancrofti

- even picograms of DNA will be detected
- Best investigation

SUB CUTANEOUS SITES

ONCHOCERCA VOLVOLUS

02:03:45

Skin Disease	Eye Disease
Onchocercoma	River Blindness
[Sub cutaneous nodules]	[by Simulium Fly / Black Fly] Microfilariae deposit in eyes of people residing near the river bed

Black Fly

Onchocercoma

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LOA LOA

02:05:55

Skin	Eye
CALABAR SWELLINGS	LOASIS
- Migratory swellings	([BY CHRYSOPS fly])

Loa loa

DRACUNCULUS MEDINENSIS [Guinea Worm]

02:07:10

- Eradicated from India
- Last case was reported from Rajasthan
- Match stick method was used to remove worm from lesion (Blister)

Match stick method for guinea worm

TRICHINELLA SPIRALIS / MUSCLE WORM

02:08:57

- Transmitted by eating larvae in uncooked pork

↓

Mature adults M & F Worms

↓

Fertilize

↓

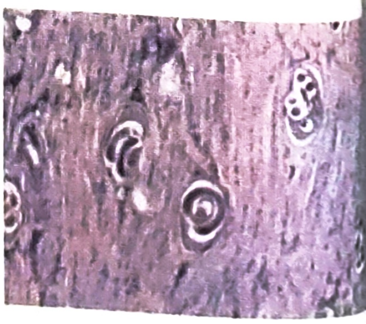
Larvae produced [VIVIPAROUS]

↓

Adult Forms

↓

Finally settle in muscles



Trichinella spiralis

02:11:05

VISCERAL LARVAE MIGRANS by
– Toxocora canis – Toxocara catis – Angiostronglus cantonensis

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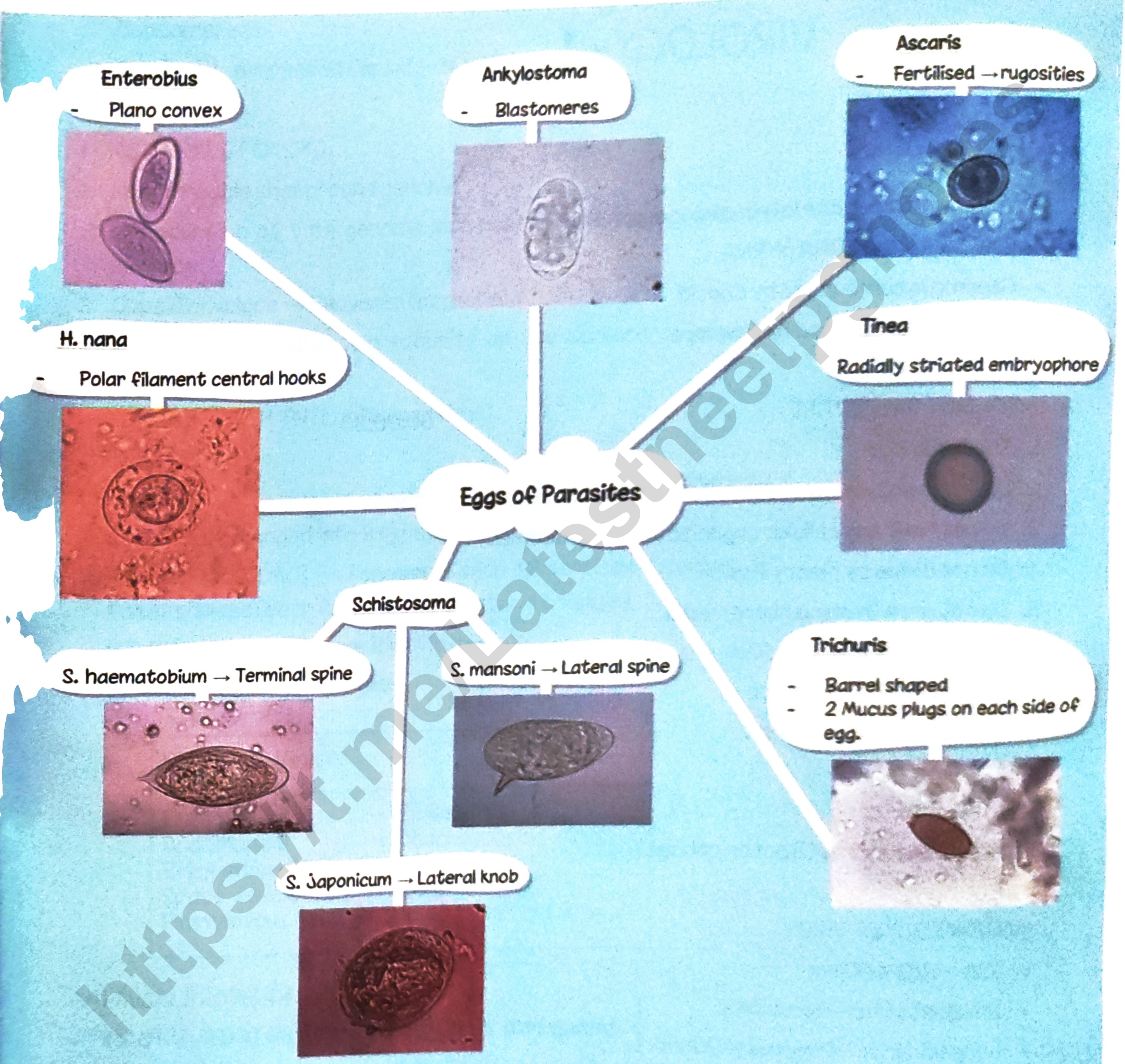

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Unit: 5 Virology

Chapter 11

VIROLOGY- 1

INTRODUCTION

VIRUSES

- Viruses are obligate intracellular organisms
- Contains either DNA or RNA
- Genome is surrounded by Capsid
- Capsid is surrounded by Envelope

IMPORTANT PROPERTIES

00:00:30

1. Smaller in Size
2. Lack the enzyme necessary for Protein & Nucleic acid synthesis
3. Do not have any cellular organization or Ribosomes
4. Do not divide by binary fission
5. Do not grow in any culture media
6. Contain either DNA or RNA
7. Resistant to antibiotics
8. Host specificity present
   1) Animal Virus
   2) Plant Virus
   3) Bacterial Virus [Bacteriophage]

MORPHOLOGY OF VIRUS

00:05:54

- Size → 20-400 nm
- Smallest virus → Parvovirus } Among DNA Viruses
- Largest virus → Poxvirus (400nm) } Among DNA Viruses
- Smallest virus (overall) → Hand, foot & mouth disease virus (RNA virus)

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STRUCTURE 00:07:22

- Central core of Nucleic acid of a Virion is called Genome
- Genome is surrounded by a protein coat called Capsid [made of Capsomeres]
- Genome & surrounded Capsid → NUCLEOCAPSID

FUNCTIONAL OF CASPID

1. Impermeable shell around genome
2. Introduction of Viral genome into host cell by adsorbing on cell surface
3. Capsid envelope → Bilayered lipoprotein

- Virus codes glycoprotein subunits on the surface of envelope [PEPLOMERS]
- PEPLOMERS – H & N [in influenza virus]

SYMMETRY 00:11:27

→ Capside is arranged into 3 types of symmetries

- ICOSAHEDRAL [MC] → Polyygonal with 12 vertices / corner, 20 facets/slides [each facet is an equilateral triangle]
- HELICAL (2nd MC) → Some RNA Viruses
- Complex (LC) → Pox Virus

Shapes of Viruses	00:14:00
Bullet Shaped	→ Rabies virus
Rod Shaped	→ Tobacco mosaic virus
Brick shaped	→ Pox virus
Space vehicle	→ Adeno virus
Filamentous	→ Marburg & Ebola virus

CHEMICAL PROPERTIES 00:16:25

- INACTIVATED AT 56°C X 30 MIN
- Stable & can be stored at - 40 to -70°C by lyophilization
- Radiation → UV rays, X Rays, Gamma rays inactivate the virus
- Lipid solvent → Chloroform, Ether & Bile salts destroy enveloped viruses easily

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- Disinfectants→ Like oxidizing agents of Chlorine, Iodine & H_2O_2 Kills most viruses
- Enteroviruses are resistant to Phenol & Chlorination

REPLICATION

00:21:44

Steps →'A PUB MR' → Mneumonic

1. Adsorption
2. Penetration
3. Uncoating
4. Biosynthesis/Assembly
5. Maturation
6. Release

ADSORPTION

- Comes in contact by RANDOM COLLISION
- Depends on presence of SPECIFIC RECEPTORS
- Eg:
  (1) HIV → Envelope proteins gp 120 / gp 41
  ↓
  Binds to CD_4 T cell receptors
  (2) Influenza Virus A
  - H → Hemagglutinin [Adsorption on Pharyngeal cells]
  - N → Neuraminidase

VIROPEXIS/PENETRATION

- VIROPEXIS → After attachment, the virus particle is taken inside the cell
- In enveloped viruses, envelop fuses with plasma membrane of host cell
  - releasing the Nucleocapsid into the cytoplasm

UNCOATING

- Physical separation of Nucleic acid from Capsid occurs
- Enveloped viruses enter cell by receptor mediated Endocytosis & lose the outer covering by action of lysosomal enzymes

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BIO SYNTHESIS

- Viral genome directs, the biosynthesis machinery of the host cell to synthesize products for "New Progeny Virus"
- Viral proteins are synthesized mostly in cytoplasm
- Most of the DNA Viruses replicate in the Nucleus except Pox virus (replicate in cytoplasm)
- Most of the RNA viruses replicate in the cytoplasm except Influenza & Retro virus (replicate in Nucleus)
- Most of DNA Viruses are DS except Reo Parvo virus
- Most of RNA viruses are SS except Reo virus
- In case of ss DNA virus (Parvo virus) → Viral nucleic acid utilizes cellular DNA polymerase for making viral DNA double Stranded
- Double stranded DNA Virus → Viral nucleic acid enters the Nucleus of host cell

– Transcription of Parental DNA of virus

↓

Early mRNA

↓ Translation

Early non- structural proteins

– Early non - structural proteins help parental DNA

↓

Shut host cell machinery

↓

Progeny DNA

– Progeny DNA undergo transcription

↓

Late mRNA

↓

Translated to Late structural proteins [Capsid & Envelope proteins]

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- In case of RNA Virus

  ss RNA Viruses are classified into 2 categories

1) The Positive sense RNA [Plus strand]

↓

Acts as mRNA

↓

Early proteins

↓

RNA Polymerase activity

Direct replication of positive ss RNA

↓

Negative ss RNA → Positive ss RNA → Late proteins are produced

- Eg. Picorna & Togavirus

2) The Negative sense [minus strand]

↓

Posses their own RNA Polymerase for mRNA transcription

↓

+ve sense RNA

↓ Transcription

mRNa

↓

Proteins

- Eg. Rhabdo, Orthomyxoviridiae, Paramyxoviridiae

- ds RNA VIRUS [Reovirus] have segmented genome

  Each segment codes for 1 Polypeptide

- + strand RNA can act as mRNA

  ↓ Translation

  Proteins

- Both + & - strands serves as templates for synthesis of complementary strands to form a duplex

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- In case of RETRO Virus,

ss RNA genome
↓ Viral Reverse transcriptase [RNA dependent DNA polymerase]
ss DNA
↓
DNA : RNA Hybrid

- Reverse transcriptase enzyme has Ribonuclease activity also by which it digest RNA from DNA : RNA Hybrid

ss DNA
(using DNA polymerase activity of same Reverse transcriptase enzyme)
↓
ds DNA
↓
Integrated into the host cell chromosome (Nucleus) by viral Integrase

- Integrated DNA serves as a template to produce mRNA
- mRNA → Early & Late proteins

ASSEMBLY

- Viral nucleic acid & proteins are packaged to form progeny viruses [Nucleocapsid]
- Assembly may takes place in host cell Nucleus or Cytoplasm
- DNA viruses mostly assemble in Nucleus except Pox & Hepadnavirus
- RNA viruses mostly assemble in cytoplasm

MATURATION

- May take place in Nucleus or Cytoplasm

Eg. In Nucleus → Herpes, Adeno Virus
In Cytoplasm → Picorna, Pox virus

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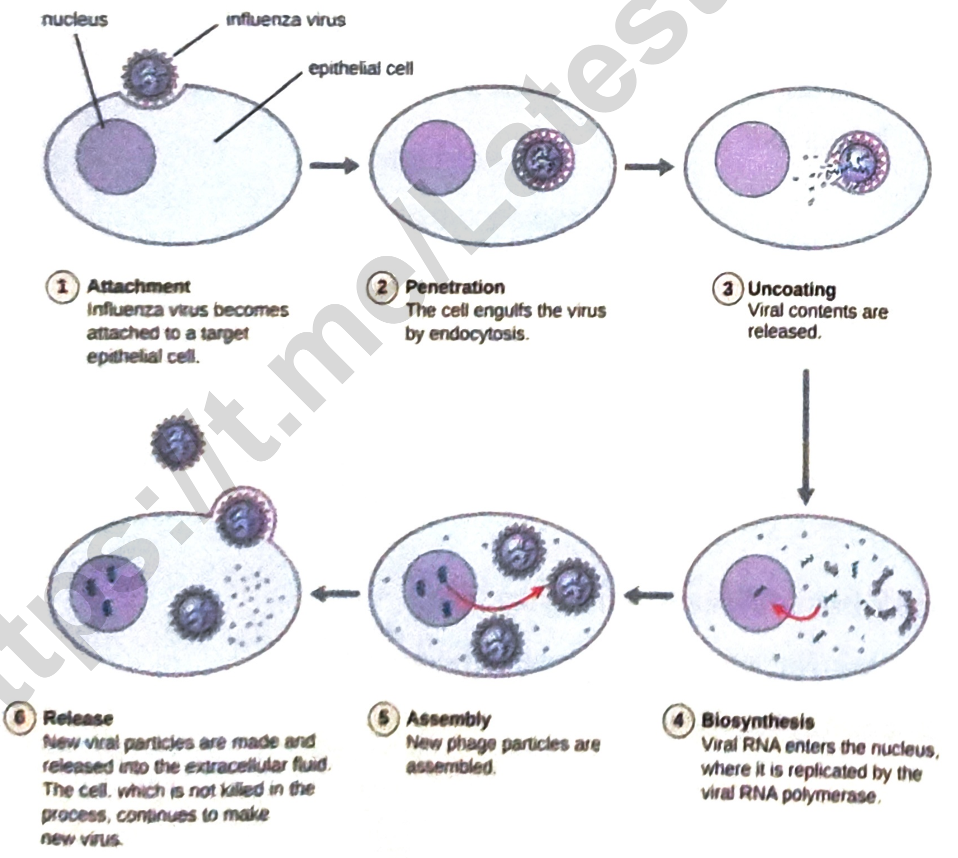
- In case of enveloped virus, the envelope is derived from nuclear membrane (if virus assemble in nucleus) or from plasma membrane during the process budding (if virus assemble in cytoplasm)

RELEASE BY

1) LYSIS → Lysis of host cell is done by non-enveloped viruses & Bacteriophages
2) BUDDING → happens in enveloped viruses - they acquire a part of host cell membrane to form Lipid part of envelope

- Eg; Envelope is acquired in influenza virus from plasma membrane & for herpes virus from Nucleus membrane
- Viral glycoproteins are inserted into the envelopes
- Excess viral glycoproteins are synthesized to saturate cell receptors so that virus will not stick to host cells following Release



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ABNORMAL REPLICATIVE CYCLES

01:16:14

INCOMPLETE VIRUSES

- Result from defective assembly [proteins are assembled without nucleic acid]
- Eg. Von Magnus Phenomenon [Influenza Virus]
  - High Hemagglutination titer but low infectivity

PSEUDO VIRUSES

- During replication of viruses, Capsid occasionally encloses Host cell nucleic acid Instead of viral nucleic acid. They are neither infective nor replicative

ABORTIVE INFECTION → d/t wrong selection of host cells by virus

MUTATION

01:21:15

- Occurs at 10-4 to 10^{-10} (10^{-8}) per base pair per generation
- Mutation can happen spontaneously or induced by 5-Fluorouracil, UV light

CONDITIONAL LETHAL MUTANTS

- Can grow only in some permissive conditions
- Eg: Temperature sensitive mutants → can grow at 28-31°C but not at 37°C → used for making influenza vaccines

REASSORTMENT

- Mostly happens in influenza virus A
- Two strains of influenza A infect a host cell

  ↓

  Gene exchange b/w them happens

  ↓

  Reassortants are formed
- Reassortmant happens in influenza due to its segmented genome (8 segments)

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VIRAL INTERFFRENCE

- Two viruses infect a host cell [one virus leads to inhibition of other virus]
- One virus can do blocking of the host cell receptor or destroys host cell receptor resulting in growth inhibition of another virus
- The 1st virus may compete with 2nd virus for replication apparatus & can induce Interferon production to kill second virus
- Eg. Rubella (Togaviridae)
  Polio (Picornaviridae)

CULTIVATION OF VIRUS

01:30:33

(1) Hen's Egg Inoculation

(2) Animal Inoculation

(3) Cell Culture / Tissue Culture

1. HEN'S EGG INOCULATION [CANDLING]

- CAM → Chorio Allantoic Membrane → Variola/Vaccinia [pox virus]
- Allantoic cavity → Vaccine production
  - Y → Yellow fever [17D]
  - I → Influenza
  - R → Rabies
- Amniotic sac → primarily used for isolation of influenza virus
- Yolk sac → C → Chlamydia
  A → Arbo Virus
  R → Ricketisiae

2. ANIMAL INOCULATION

- Newborn suckling mice [<48 hr] → used for proper cultivation of Arbo virus, Coxsackie Virus
- To study Viral oncogenesis → Hamsters, Guinea pigs, Rabbits, Monkeys are used
- ROUTES OF INOCULATION
  - Intracerebral
  - Intranasal

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- Intraperitoneal
- Sub cutaneous

- Animals are observed for the signs of illness & stained for inclusion bodies

3 Cell CULTURE / TISSUE CULTURE

1. Organ culture → Small bits of organs [Tracheal ring - Corona Virus]
2. Explant culture → Fragments of minced tissue
3. Cell culture → Mostly done now a days

CELL CULTURE LINES [Growth medium]

1. PRIMARY CELL LINES

- obtained from fresh organs of animals or humans
- allow only 5-10 divisions
- Primarily used for isolation of virus
- Eg.
  - Rhesus monkey Kidney cell lines
  - Chick embryo fibroblast cell lines

2. SEMI CONTINOUS / DIPLOID CELL LINES

- usually Fibroblasts
- allow 10-50 divisions
- can be used for longer storage of virus
- Eg. Human Fibroblast cell lines (WI-38)

3. CONTINUOUS / CANCER CELL LINES

- Serially subcultured
- unlimited divisions
- Eg.
  - Hela → derived from Cervical cancer of a lady
  - Hep 2 → derived from Human Epithelioma of larynx
  - BHK → derived from Baby Hamster Kidney
  - Vero → derived from Vervet Monkey Kidney cell line

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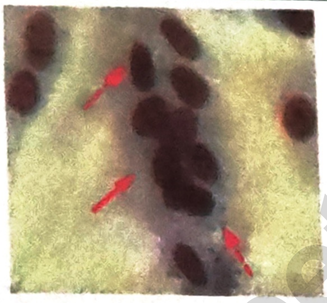
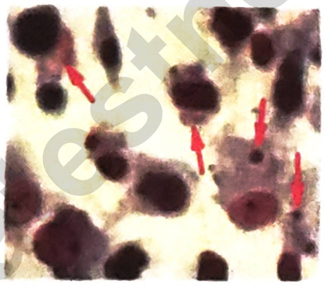
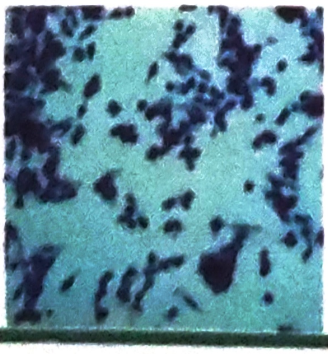
CYTOPATHIC EFFECT

01:48:00

→ helps in identification of Virus

- Multinucleate Giant cell formation (Syncytium formation)
  - Measles Virus/ RSV
- Cell necrosis & lysis → Enteroviruses
- Cell clumping [grape like clusters] → Adenovirus
- Cytoplasmic vacuolation → SV - 40

Syncytium and faint basophilic cytoplasm inclusion bodies	
Pink eosinophilic cytoplasmic inclusion bodies (arrows) and cell swelling	
Cytoplasmic stranding (arrow) and nuclear inclusion bodies (drashed arrow)	
Cell enlargement, rounding, and distinctive" grape-like" cluster	

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INCLUSION BODIES

01:50:33

- Replicating aggregates of the virus either intranuclear or intracytoplasmic
- Can be seen with normal power microscope

INTRA CYTOPLASMIC INCLUSION BODIES [Eosinophilic]

1) Negri/bodies → Rabies
2) Guarnieri bodies → Vaccinia
3) Paschen bodies → Variola
4) Bollinger bodies → Fowl pox
5) [Henderson Peterson] Molluscum bodies → Mulluscum contagiosum

INTRA NUCLEAR INCLUSION BODIES

1) Cowdry type A inclusions seen in

   H → Herpes virus [Lipschvitz bodies also seen]

   A

   Y → Yellow Fever virus [Torres bodies also seen]

2) Cowdry type B inclusions are more circumscribed & seen in

   B

   A → Adeno virus

   P → Polio virus

- Intra nuclear + Intra cytoplasmic Inclusion Bodies Seen In
  - Measles [aka WARTHIN FINKELDAY GIANT CELLS]
- Intra nuclear + Intra cytoplasmic inclusion bodies also seen in
  - CMV [Owl eye appearance of infected cells]

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DNA VIRUSES

HERPES

FAMILIES

01:57:47

α VIRINAE	β VIRINAE	γ VIRINAE
→ Includes HSV1 HSV2 HHV3 → Affects Ganglionic cells	→ Includes HHV 5 HHV 6 HHV 7 → Affects salivary glands	→ Includes HHV 4 HHV 8 → Affects B cells (d/t CD_{21}/CR_2 Receptors over B cells)

- HHV - 3 → Varicella zoster [chicken pox virus]
- HHV - 4 → EBV
- HHV - 5 → Cytomegalo virus
- HHV - 8 → Kaposi Sarcoma causing virus
- HSV - 1 → Herpes simplex virus 1
- HSV - 2 → Herpes simplex virus 2

HSV

- HSV 1 → Causes infection above the waist — not a rule
- HSV 2 → Causes infection below the waist

Herpes labialis

MUCOSAL INVOLVEMENT

- mc site → Buccal mucosa
- Causes → Acute Gingivo stomatitis
- mc recurrent herpes lesion → Herpes labialis

Eczema Herpeticum by HSV 1/2

CUTANEOUS INVOLVEMENT

- mc site → Face
- HERPETIC WHITLOW → Occurs in Nurses, doctors
- HERPETIC GLADIATORUM → Occurs in Wrestlers

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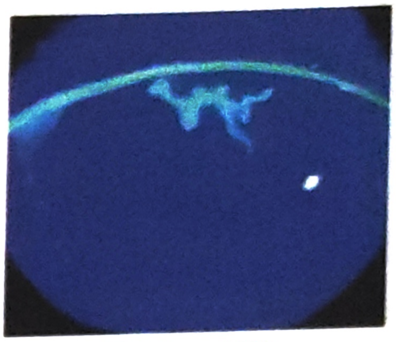
CNS

- Temporal lobe encephalitis
- Mollaret's Meningitis [Recurrent lymphocytic meningitis]

OPHTHA INVOLVEMENT

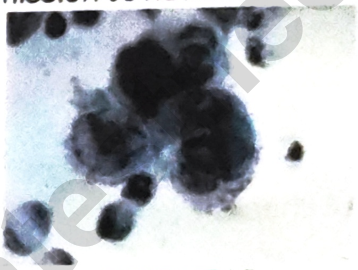
- Cause acute dendritic ulcers
- Steroids are contraindicated
- Topical Acyclovir indicated



HSV Keratitis

GENITAL INVOLVEMENT

- causes B/L Vesicular, Pustular, Erythematous lesions
- Pregnant female with HSV2 infection near term
  - Cesarean section done to avoid transmission to newborn



TZANCK CELLS

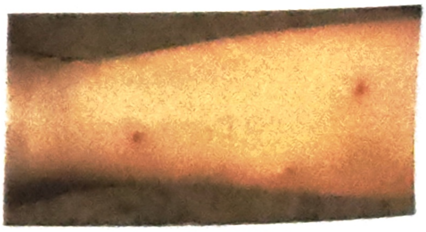
LAB DIAGNOSIS

1 PCR } For HSV1 & 2
2 ELISA }
3 TZANCK SMEAR

Tissue biopsy + Toluidine blue → TZANCK CELLS [Multinucleated giant cells]

VARICELLA ZOSTER/CHICKEN POX VIRUS 02:10:43

- Portal of entry → Respiratory tract / conjunctiva
- SAR (secondary attack Rate) → >90%
- Infectivity period → 2 days before ←RASH→ 5 days after [until the scab Fails]
- Centripetal Rash +nt, DEW DROP APPEARANCE & Pleomorphism seen
- IP → 14-16 days
- MC complication → Secondary bacterial infections
- MC extra cutaneous involvement → Brain
- Most serious complication → Varicella Pneumonia



CHICKEN POX

CHICKEN POX

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- CONGENTIAL VARICELLA SYNDROME
- Chorio retinitis
- Limb hypoplasia
- Microcephaly
- Cicatrizing lesions
- Vaccine strain → OKA Strain

LAB Diagnosis

1. PCR/ELISA For HHV -3
2. TZANCK SMEAR

REACTIVATION → Herpes Zoster [Shingles]

- Occurs in stress
- Types

1. HZ Oticus/RAMSAY HUNT SYNDROME
   - Bell's palsy
   - Vesicles over EAC, TM & cheeks
   - Geniculate ganglion is involved

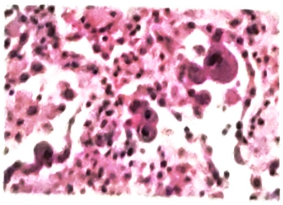
HZ Ophthalmicus

2. HZ Ophthalmicus
   - Single sided lesion +nt on Face
   - Dermatomal involvement [T3-L3]
   - Ophthalmic branch of Trigeminal nerve is involved

- LAB DIAGNOSIS → PCR/ELISA available

CMV 02:21:20

- Largest Herpes virus
- OWL EYE APPEARANCE of infected cell seen



CMV OWL EYE APPEARANCE

Congenital CMV SYNDROME

FEATURES

- Hepatosplenomegaly [mc]
- Petechial lesions

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- Chorio retinitis
- Calcifications
- Convulsions
- Microcephaly
- Mental retardation

MONONUCLEOSIS LIKE SYNDROME

FEATURES

- Atypical lymphocytes present [CD_8 T cells mainly]
- Paul Bunnel Test → -ive
- Causes CMV Pneumonia in Post Kidney transplant, Post bone marrow transplant [Post transplant 1-4 months are more vulnerable to develop CMV pneumonia]

CMV RETINITIS

- Risk Factor → CD_4 count → <50 (HIV +ve patient)
- DOC → GANCICLOVIR

LAB DIAGNOSIS

1. CULTURE on Human Fibroblast cell line [WI-38]
2. PCR For CMV
3. ELISA For CMV

EBV

- Affects B cells → Leads to Polyclonal proliferation of B cells

↓

Number of Cancers

- DISEASES

1. NASO PHARYNGEAL CA [NPC]
2. BURKITT'S LYMPHOMA [Starry sky appearance]
3. HODGKIN & NON HODGKIN'S LYMPHOMA
4. ORAL HAIRY LEUKOPLAKIA [OHL]
5. DUNCAN SYNDROME → X linked lympho proliferative syndrome

BURKITT'S LYMPHOMA
(Tumor of Jaw)

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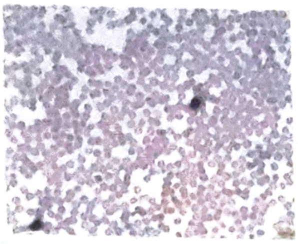
6. TONSILLAR CARCINOMA

7. INFECTIOUS MONONUCLEOSIS/KISSING DISEASE

- Fever, Rash, Lymphadenopathy
- CD_4: CD_8 Ratio reverses for sometime & then comes back to normal
- Atypical lymphocyte +nt
- Paul Bunnel test is positive



Atypical lymphocytes

LAB DIAGNOSIS

1. PCR For EBNA [EPSTEIN Barr Nucleic acid]
2. PCR For VCA [Viral Capsid Antigen]
3. ELISA For EBV

Kaposi Sarcoma

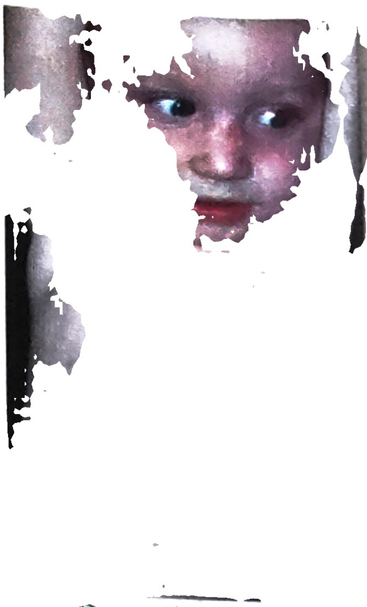
Kaposi Sarcoma Lesions

HHV – 8 [Kaposi Sarcoma causing virus] 02:34:56

- Common among HIV +ive
- Vascular tumor
- also a/w Primary effusion lymphoma & Castle man's disease

HHV 6 & 7 02:36:25

- Causes Exanthem subitum / Roseola infantum

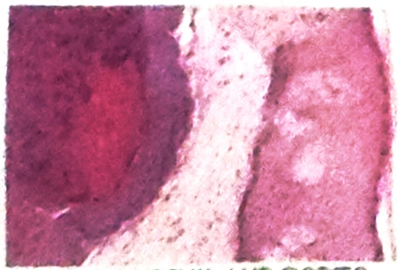


Roseola infantum

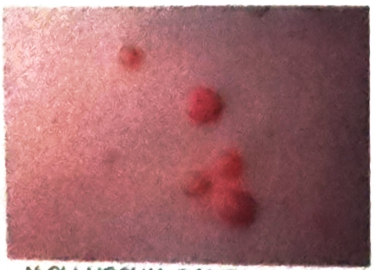
POX VIRUS 02:37:20

MOLLUSCUM CONTAGIOSUM (MCV)

- Pearly white umblicated nodule over the skin/genitals +nt
- Caused by MCV 1,2,3,4 [MCV 1 → mc]
- On biopsy, HENDERSON PETERSON [HP] BODIES /MOLLUSCUM BODIES seen [Intra cytoplasmic inclusion bodies]
- Rx → PODOPHYLLIN RESIN + CRYOSURGERY



MOLLUSCUM / HP BODIES



MOLLUSCUM CONTAGIOSUM

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PrepLadder

PARVO VIRUS

02:40:34

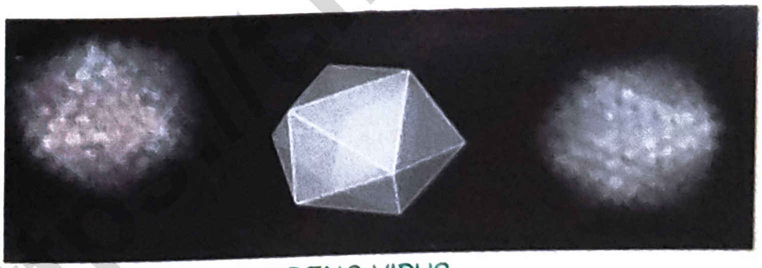
- ss DNA VIRUS
- PARVO B 19 → important serogroup
- AFFECTS ERYTHROID PRECURSOR CELLS
  - APLASTIC CRISIS in Sickle cell anemic Patients
- ASSOCIATED WITH
  - NIHF [Non Immune Hydrops Fetalis]
  - Polyarthralgia syndrome
    - Pain is more in upper limb joints than Lower limb joints
  - Erythema infectiosum / 5th Disease
    - SLAPPED CHEEK APPEARANCE +NT

SLAPPED CHEEK APPEARANCE

ADENO VIRUS

02:44:38

- ds DNA Virus
- Non enveloped virus
- Looks like space vehicle
- Type 3,4,7,14 → Causes Pharyngo conjunctival fever or →Swimming pool conjunctivitis
- Type 8,19,37 → Causes Epidemic Kerato conjunctivitis
- Type 37 → Causes STD
- Type 40,41 → Causes Infantile diarrhea
- Type 11,21 → Causes Hemorrhagic cystitis



ADENO VIRUS

AAV & VACCINIA [AAV → Adeno Associated Virus]

- Used for making recombinant vaccines using genetic engineering

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POLYOMA & PAPILOMA VIRUS

POLYOMAVIRIDAE

- Mostly oncogenic in Lab animals
- Non - enveloped
- Icosahedral symmetry
- 45 nm in Size
- ds DNA Genome
  - Early region → Interacts with P_{53} & Rb genes
  - Late region → Capsid formation

JC VIRUS

02:51:15

- Cultured in human Fetal glial cells
- infection → Ubiquitous
- Risk Factor → HIV + ive, Post transplant, HodgKin's Lymphoma

JC virus crosses Blood Brain Barrier

↓

Infects Oligodendrocytes & Astrocytes

↓

Causes Subacute Demyelinating disease

↓

PROGRESSIVE MULTIFOCAL LEUKOENCEPHALOPATHY (PML)

- LAB DIAGNOSIS
  - PCR on brain biopsy tissue For JC virus
- No treatment, Fatal disease

BK Virus

02:54:18

- Post Kidney transplant infection [Borrowed Kidney]
- Cultured in human diploid cell lines
- Latency in Kidney & reactivated by Kidney transplants [BK NEPHROPATHY]
- DECOY CELLS are seen in BK nephropathy

DECOY CELL

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PrepLadder

- **LAB DIAGNOSIS**
- Urine → Culture
  → PCR
- **TREATMENT** → Immunosuppression to be corrected

SV – 40 [SIMIAN VACOULATING – 40] 02:56:50

- Causes Progressive multifocal leukoencephalopathy in Rhesus monkeys
- Cytopathic effect → Cytoplasmic vacoulation

Merkel cell carcinoma virus

KI Virus [Karolinska institute virus]

WU Virus [Washington university virus]

PAPILLOMAVRIDAE 02:59:00

1. All are oncogenic in Lab animals
2. Non – enveloped
3. Icosahedral symmetry
4. ds DNA Genome [Circular]

- Regulatory region
- Late region → L1 & L2 → Capsid
- Early region → E1, E2, E3, E4, E5, E6, E7
  - E6 → Inactivates products of P53 gene
  - E7 → Inactivates products of Rb gene

INFECTIONS CAUSED BY PAPILOMA VIRUS

- Cause infections in the breaks of skin & mucous membrane, exposing the basement membrane

TYPES OF PAPILLOMA VIRUS ON NUCLEOTIDE SEQUENCING

›200 Genotypes

- Cutaneous
  - Low Risk: 1,2,3
  - High Risk: 5,8
- Mucosal
  - Low Risk: 6,11
  - High Risk: 31,33,16,18

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PrepLadder

DISEASES CAUSED BY PAPILLOMA VIRUS

1. SKIN WARTS

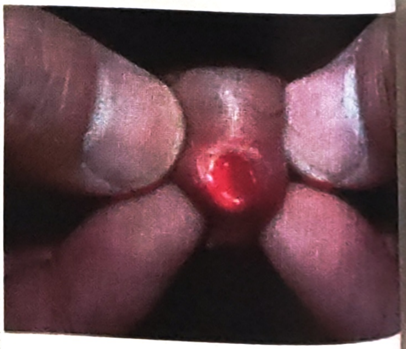
- Most common types → Type 1 to 8
- Epidermoplasia verruciformis / Tree Man syndrome caused by HPV 5,8

Tree Man syndrome

2. GENITAL WARTS

→ Condyloma acuminata
→ Laryngeal warts

} Caused by HPV 6,11



Condyloma Acuminata

3. CARCINOMA

- FEMALES → Cervical Ca, Vulval, Vaginal, Anal Ca
- Males → Anogenital carcinoma [Penile, Anal]

VACCINES

a. CERVARIX [Bivalent vaccine] → HPV 16,18
b. GARDASIL [Quadrivalent vaccine] → HPV 6,11,16,18
c. GARDASIL -9 → HPV 6,11,16,18,31,33,45,52,58

DIAGNOSIS

HISTOPATHOLOGY

- Koilo cytosis
- Cytoplasmic vacuolation along with Nuclear enlargement

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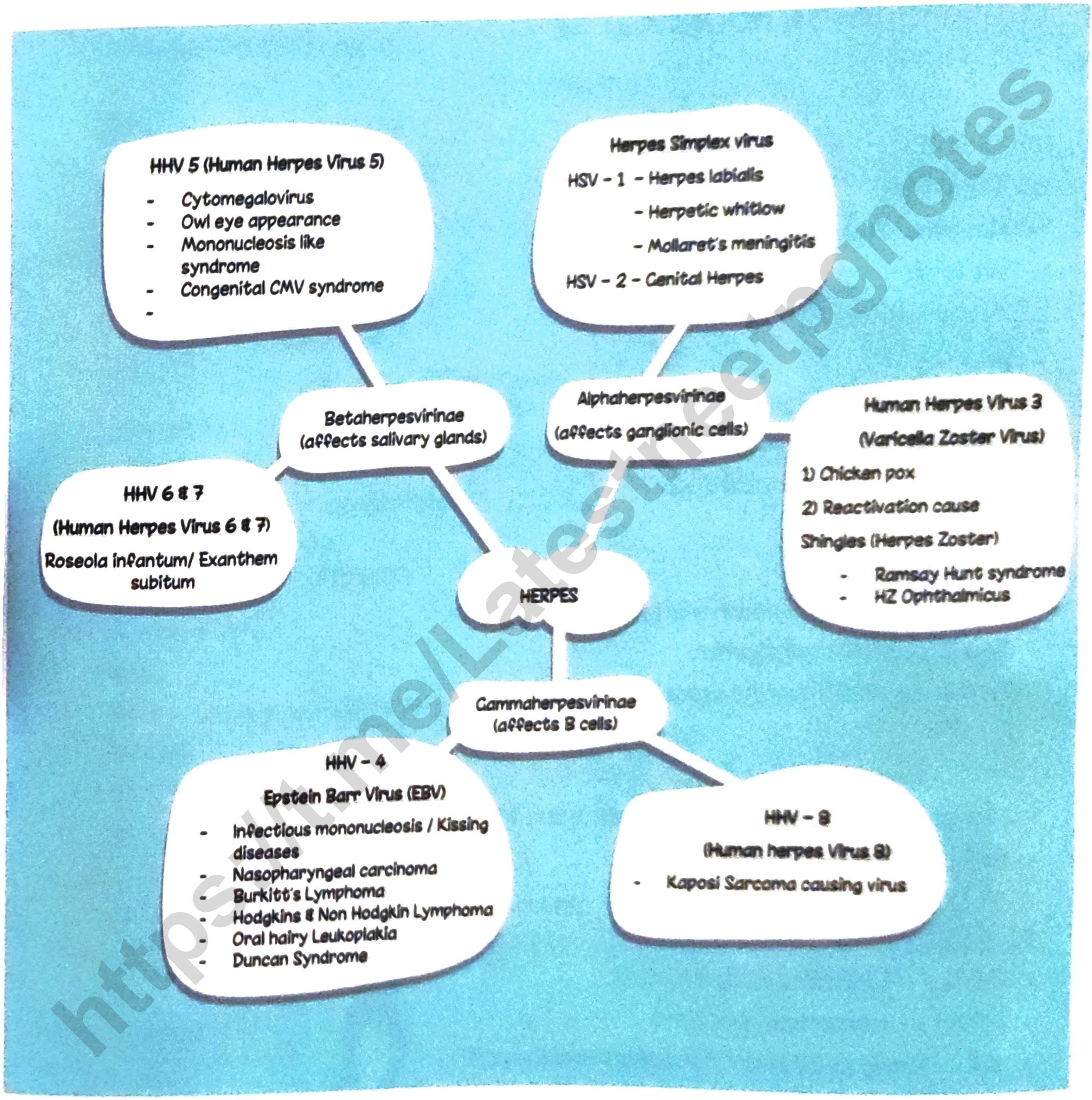

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PrepLadder

Chapter 12

VIROLOGY 2

RNA VIRUSES

PICORNAVIRIDAE

00:00:13

Includes small RNA Viruses

1. Polio Virus
2. Coxsackie virus
3. Entero virus
4. ECHO virus [Entero cytopathogenic Human orphan Virus]
   - Causes Aseptic meningitis
5. Rhino Virus → Common cold virus

POLIO VIRUS

00:03:14

Type 1 → Most common wild type

Type 2 → Most antigenic

Type 3 → VAP [Vaccine associated Paralysis]

SWITCH OVER

- New vaccines are Bivalent from old vaccines [Trivalent]
- No Type 2 Strain

POLIO

- Route → Faeco oral contamination
- >95% → Inapparent infections
- <1% → Paralysis (Anterior horn cells are damaged)

Polio

- Tonsillectomy should be avoided during a polio epidemic
- More physical activity should be avoided during polio epidemic

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Lab Diagnosis

- CULTURE → on Monkey Kidney cell lines
- ELISA / PCR For Polio virus
- ANTIGENS
  - C antigen [Coreless antigen] } Type specific
  - D antigen [Dense antigen] } Type specific

COXSACKIE VIRUS

00:09:33

COXSACKIE VIRUS A

- Causes
  - Flaccid Paralysis in mice
  - Herpangina [Acute Vesicular pharyngitis]
  - Acute Hemorrhagic Conjunctivitis [Cox A_{24} + Enterovirus – 70]
  - HFMD [Hand Foot Mouth Disease] → COX A_{16}
  - Aseptic meningitis

Herpangina

COXSACKIE VIRUS B

- Causes
  - Bornholm's disease [Acute Pleurodynia or Devil's grip]
  - DM [B4]
  - HFMD
  - Aseptic meningitis
- Inoculation into mice cause Spastic paralysis
- CULTURE → On New born Suckling mice [<48 hrs]

*Arbo viruses are also cultured on new born suckling mice

Hand, Foot & Mouth Disease

ENTEROVIRUS

00:15:33

- Includes
  - Enterovirus 70 → Causes Acute Hemorrhagic conjunctivitis
  - Enterovirus 71 → Causes HFMD
  - Enterovirus 72 → Hepatitis A

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MYXOVIRUSES

00:17:38

ORTHOMYXOVIRIDAE	PARAMYXO VIRIDAE
Includes – Influenza A,B,C	Includes - Para Influenza viruses - Mumps - Measles - Rubella (Togaviridae family, Arbovirus) - RSV - Metapneumo virus - Nipah virus (Henipa virus)

ORTHOMYXO VIRIDAE

00:20:12

INFLUENZA A, B, C

- ss RNA
- Negative sense
  ↓ RNA polymerase
- Positive sense
- Have segmented genome
  A-8 segments
  B-8 segments
  C-7 segments
- Glycoprotein H (Hemagglutinin)
  - Adheres to cell
- Glycoprotein N (Neuraminidase)
  - Receptor Destroying Enzyme
  - Process is known as ELUTION

SEGMENTED GENOME
B→Bunya virus
I→ Influenza virus
R→Reo virus
A→Arena virus

ANTIGENIC SHIFT

- Abrupt, drastic & discontinuous process
- d/t Reassortments
- Causes Pandemic
- H3N2 → H1N1

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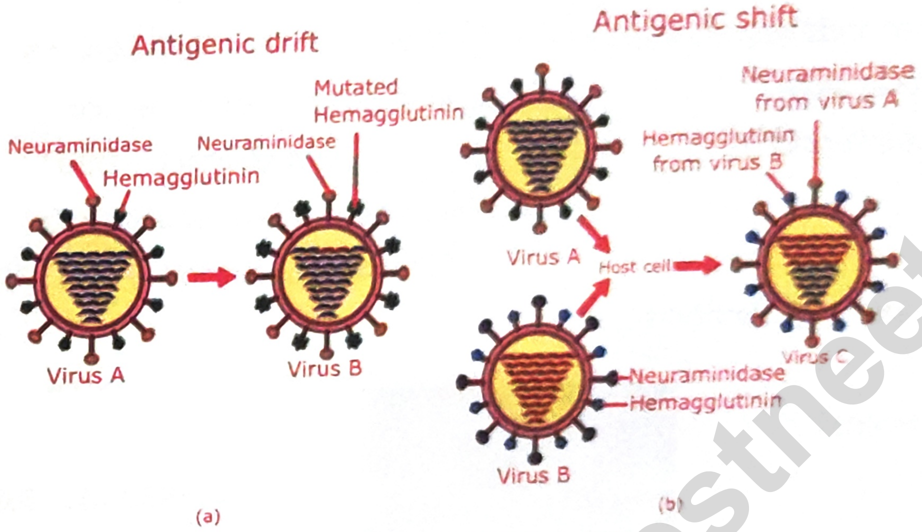
ANTIGENIC DRIFT

- Single point mutation & Continous in nature
- $H_3N_2 \rightarrow H_{3a}N_{2b}$
- Antigenic shift & drift
  - Mostly happens in Influenza A
  - Sometimes in Influenza B



ANTIGENIC SHIFT & DRIFT

H5N1 [BIRD FLU]

- Infected birds transmit infection to humans
- Human to human transmission not seen
- PB_1F_2 gene → Target the Mitochondria of cell & damages it

H_1N_1 [SWINE FLU]

- Combination of Human influenza strain, Swine strain, Avian flu strain
- LAB DIAGNOSIS
- NASO PHARYNGEAL SWAB
  - Taken with proper precautions
  - Extract RNA [with RNA Extraction Kits]

↓

Real time PCR [<2 hrs]

(RNA → cDNA → amplify)

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PrepLadder

- Chemoprophylaxis → TAMIFLU [OSELTAMIVIR], given in initial stages Of disease / during an Epidemic
- Killed & live vaccines are available → Temporary immunity

PARAMYXO VIRIDAE

00:35:07

PARA INFLUENZA

- Para influenza type 1 & 2 causes CROUP [Acute larynogotracheo bronchitis]
- Para influenza type 3 causes LRTI [Lower respiratory tract infection]

MUMPS

00:36:38

- mc presentation of Mumps → B/L Parotitis
- COMPLICATIONS
  - U/L Orchitis [in young males]
  - Oophritis [in Females]
  - Aseptic meningitis
  - Pancreatitis
- SAR → ›85%
- IP → 2-3 wks
- LAB DIAGNOSIS → PCR / ELISA for Mumps virus
- Vaccine strain → Jeryl Lynn Strain

MUMPS

*Sometimes Meningo encephalitis precedes Parotitis in Mumps infection

MEASLES

- Most characteristic feature → KOPLIK SPOT [opp. to lower 2nd Molar]
- SAR → ›90%
- IP → 10-14 days
- Infectivity period → 4 days before ← RASH → 5 days after
- Inclusion Bodies → WARTHIN FINKELDAY GIANT CELLS

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- Clinical Features
  - Diarrhoea
  - Sinusitis
  - Pneumonia
  - Otitis media

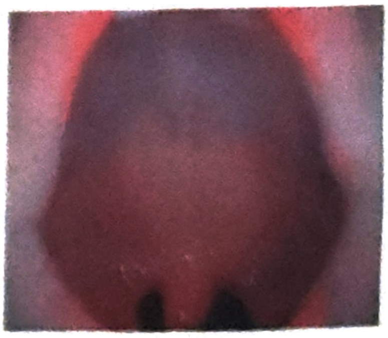
Measles Rash

- Chronic / Late (after years) complication
  - SSPE [Subacute Sclerosing Pan Encephalitis]
- Vaccine strain → Edmonston Jagreb/ Moraten/ Schwartz
  - Vaccine is given by subcutaneous route
    [BCG → Intra dermal]
  - Constituted by Distilled water
    [BCG → Normal saline]

- LAB Dx → PCR /ELISA For Measles virus

RUBELLA / GERMAN MEASLES

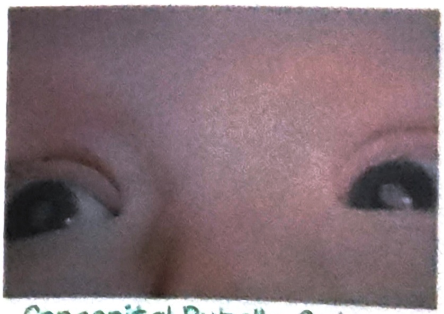
- Togaviridae family (Arbo virus)
- Infectivity period → 1 Week before ← RASH → 1 week after
- IP → 2-3 weeks
- FORCHEEIMER SPOTS are seen over Uvula
- BLUE BERRY MUFFIN RASH seen



Forcheeimer Spots

Blue Berry Muffin Rash
IUGR
Thrombocytopenia
} BLUE BERRY MUFFIN SYNDROME

- CONGENITAL RUBELLA SYNDROME
  - Classical triad
    C → Cataract
    H → Heart Disease [PDA]
    D → Deafness [sensorineural]



Congenital Rubella Cataract

- LAB DIAGNOSIS → PCR /ELISA for Measles virus

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PrepLadder

RSV Respiratory Synctial virus

00:52:03

- Causes Acute Bronchiolitis
- DOC → RIBAVIRIN
- Age group → 1 month to 6 months
- Fusion Proteins +nt → Multinucleate giant cells
- Lab Diagnosis → PCR/ELISA For RSV

METAPNEUMO VIRUS

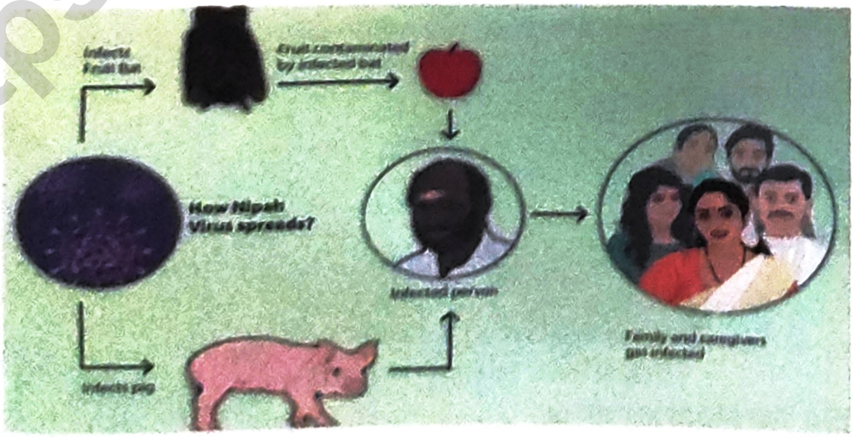
00:53:42

- Age group → 6 Months to 12 months
- causes Acute Bronchiolitis
- DOC → RIBAVIRIN
- LAB DIAGNOSIS → PCR For Metapneumo virus

NIPAH VIRUS [HENIPA VIRUS]

00:54:45

- NIPAH → Malaysian village
- Reservoir → Fruit Bats
- Amplifiers → Pigs
- MOT → Contact with the secretions of infected person
  → Eating & drinking contaminated food & water with infected secretions
  → Ingestion of contaminated Coconut palm syrup
- Mortality → > 70%
- Lab Diagnosis → PCR For NIPAH virus



NIPAH virus

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PrepLadder

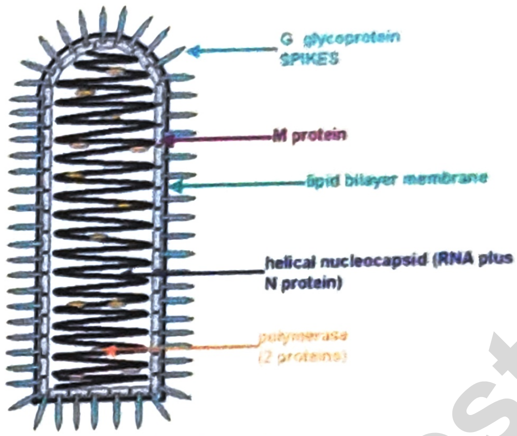
00:58:15

RABIES VIRUS

- Family → Rhabdoviridae
- Sub Family → Lyssa viridae
- ss RNA Virus (Negative sense)

  ↓ RNA Polymerase

  Positive sense
- Speed of Rabies virus → 3 mm/hr
- ↓ Ach at various sites & induce Neural cell apoptosis



Rabies virus

NATURAL / STREET VIRUS	FIXED / DUMB VIRUS
• Causes Natural / Furious Rabies • Hydrophobia } Present • Negri bodies } Present	• Causes Dumb Rabies • Used in vaccine production • Hydrophobia & Negri bodies Absent

TYPES OF BITES

CLASS I BITES

- Licks over intact skin → Nothing to be done

CLASS II BITES

- Licks over broken skin
- Slight grazing present
- No oozing of blood

} PEP

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CLASS III BITES

- Facial Bites
- Constant animal after bite
- Wild animal bites

} PEP + Immunoglobulin

Post Exposure Prophylaxis (PEP)

- Day 0, 3, 7, 14, 28
- IM route of administration
- Watch out dog/ animal for 10 days

Pre Exposure Prophylaxis

- Day 0, 7, 21 or 28
- Indicated for Veternarians, Researchers

VACCINES

- PCEC (Purified Chick embryo cell vaccine)
- PVC (Purified Vero cell vaccine)
- HDC (Human Diploid cell vaccine)

ANTE MORTEM DIAGNOSIS

- Done with Corneal smear, Nape of neck hair follicle

  ↓

  Rabies antigen demonstration by DFAT

*DFAT → Direct fluorescent antigen testing

- By Animal inoculation
- Now a days PCR is done

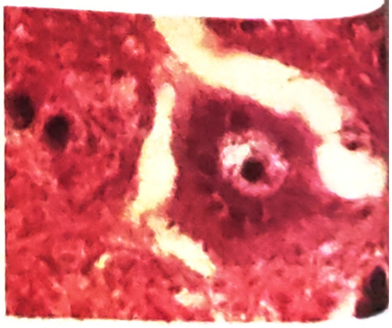
POST MORTEM DIAGNOSIS

- Demonstration of Negri bodies in Hippocampus
- Brain Biopsy tissue + Basic Fuchsin → Seller's technique

  ↓

  Negri bodies can be demonstrated

- By Animal Inoculation



NEGRI BODIES

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- NEGRI BODIES- Intra cytoplasmic Eosinophilic Inclusion bodies
- DOSE OF RABIES IMMUNOGLOBULIN – Human Rabies Ig (20 units / kg)

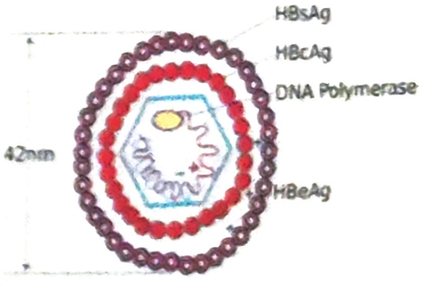
HEPATITIS VIRUS

01:14:03

HEP A	HEP B	HEP C	HEP D	HEP E
Picorna Viridae	Hepadnaviridae (only DNA virus among all Hepatitis virus)	Flaviviridae	Dependent over Hep B	Calciviridae
Faeco oral transmission	Parenteral, Sexual & Vertical transmission	Parenteral (mainly)		Faeco oral transmission
IP →15-50 D	IP→50-150 D	IP→15-150 D	IP→50-150 D	IP→15-50 D
Fulminancy 0.1% (adults)	0.1%-1%	0.1%	5-20%	1-2% →20% (pregnancy)
Chronicity	1-10%	>85%		
Carrier	0.1-30%	1.5-3.2%		

HEPATITIS A (aka Enterovirus 72)

- Only Hepatitis virus that can be cultured→has good vaccine
- LAB DIAGNOSIS
  - ELISA →IgM anti HAV demonstration
    - Stool examination
    - Serum examination [more significant]



HEPATITIS B VIRUS

HEPATITIS B

- FORMS
  - Spherical → most common
  - Tubular
  - Done particle → complete virus genome [42 nm]

MORPHOLOGICAL FORMS OF HBV

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- ACUTE HEP B

- HBs Ag + IgM anti HBC → markers for Acute Hep B
- IgM anti HBC [c-core Ag] → Better marker for depicting Acute Hep B
- HBe Ag +ive → Active Replicating phase
  → Maximum liver damage occurs
- Anti HBe +ve → Active replicative phase → ↓ing

- CHRONIC HEP B → >6 months HBS Ag +ive
  - Chronicity → 1-10%
  - Carrier Rate → 0.1 - 30 %

- If a person is vaccinated,

- Anti HBs should be > 10 IU / ml
- Schedule → 0, 1, 6 months
- Hep B vaccine
  - Recombinant vaccine
  - Prepared in yeast

- GENES

  P → Polymerase [largest gene]

  C → Precore/ Core

  S → Surface Antigen

  X → Carcinogenicity

- Pre core mutants → lack HBe Ag & carry poor prognosis
- Escape mutants → no HBs Ag

LAB DIAGNOSIS

- PCR For HBV DNA [series of PCR done]
- Hepa card Test For HBs Ag

HEPATITIS C

- MAXIMUM CHRONICITY → >85%
  - Associated more with
    - Cirrhosis
    - Hepatocellular carcinoma

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- No Good vaccine available
  - d/t QUASI SPECIES [Antigenic variations]
- LAB DIAGNOSIS
  - Series of PCR For HCV
  - ELISA
  - HCV TRIDOT Testing

HEPATITIS D

1) Co-infection → Hep B+D [reduce each other's virulence]
2) Super infection → B^D [more dangerous ; 5-20% Fulminant]

- LAB DIAGNOSIS → ELISA For IgM anti HDV

HEPATITIS E

- Fulminancy → 1-2%
  → 20% [in pregnancy]
- LAB DIAGNOSIS → ELISA For IgM anti HEV [Serum > Stool]

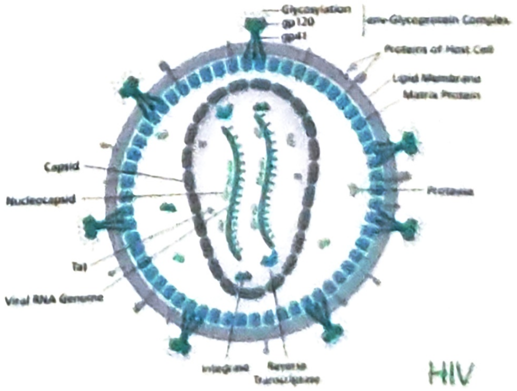
NON-ENVELOPED RNA VIRUSES

P → Picornaviridae → Hepatitis A
A → Astroviridae
R → Reoviridae
C → Calciviridae → Hepatitis E

HIV

01:44:22

- Family → Retroviridae
  Sub Family → Lenti virinae



HIV

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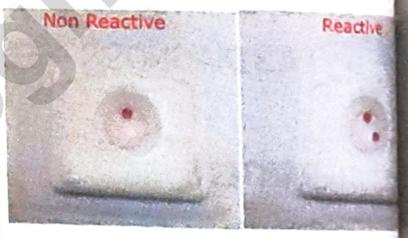
- Prevalence in India
  - HIV 1 >>> HIV 2
  - in HIV 1 → group 'M' → sub type 'C'

- STRUCTURAL GENES OF HIV 1

gag → codes For Capsid, Matrix protein, p24, p17, p18
env → codes For Gp 120 / Gp 41
pol → codes For p31, p51, p66
(Polymerase / Reverse transcriptase)

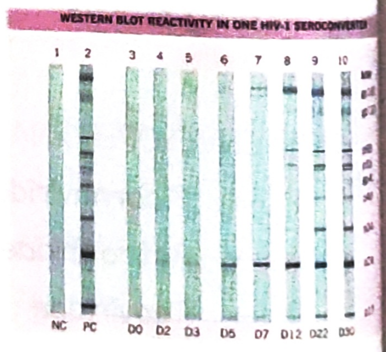


HIV Tri-Dot Test

- LAB DIAGNOSIS
  - Most sensitive test for HIV → ELISA
  - Most specific test for HIV → HIV RNA PCR > Western Blot

- MODES OF TRANSMISSION [MOT]

1) Sexual transmission → 0.1-1% [mc MOT in India]
2) Blood Transfusion → >90%
3) Needle stick injury → 0.5-1% [0.3-3%]
4) Mother to child transmission → 30%


WESTERN BLOT ACTIVITY IN

- Can ↓ transmission from Mother to child
  - to 15% → in developing countries
  - to 0-5% → in developed countries by avoiding breast Feed

NACO STRATEGIES

STRATEGY I	→	done in Blood banks
STRATEGY II A	→	done by PSM [For HIV Surveillance]
II B	→	done in Symptomatic HIV persons
STRATEGY III	→	done in Asymptomatic persons

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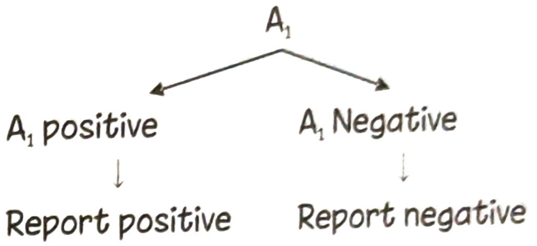
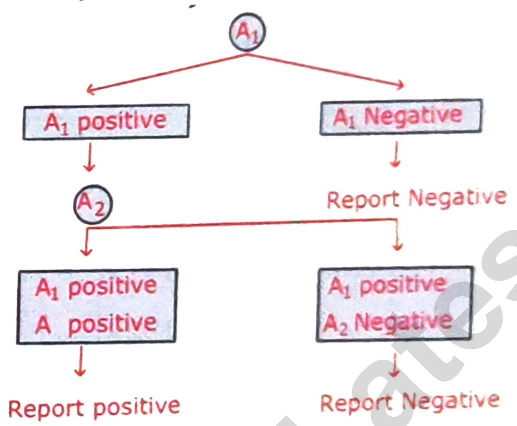
STRATEGY I / ALOGRITHIM I

- Done for Transfusion & Transplantation safety
- One test Kit is required



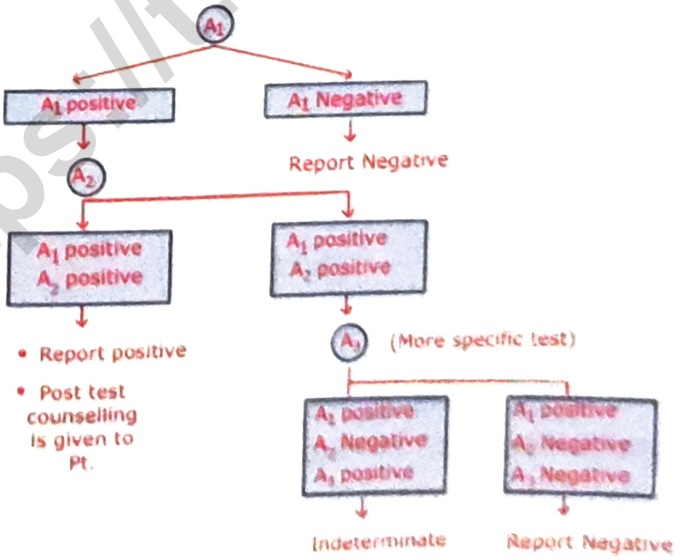
STRATEGY II A / ALOGRITHIM II A

- Done for Surveillance by PSM department
- 2 test Kits are required



STRATEGY II B / ALOGRITHIM II B

- done in Symptomatic individuals
- 3 test Kits required



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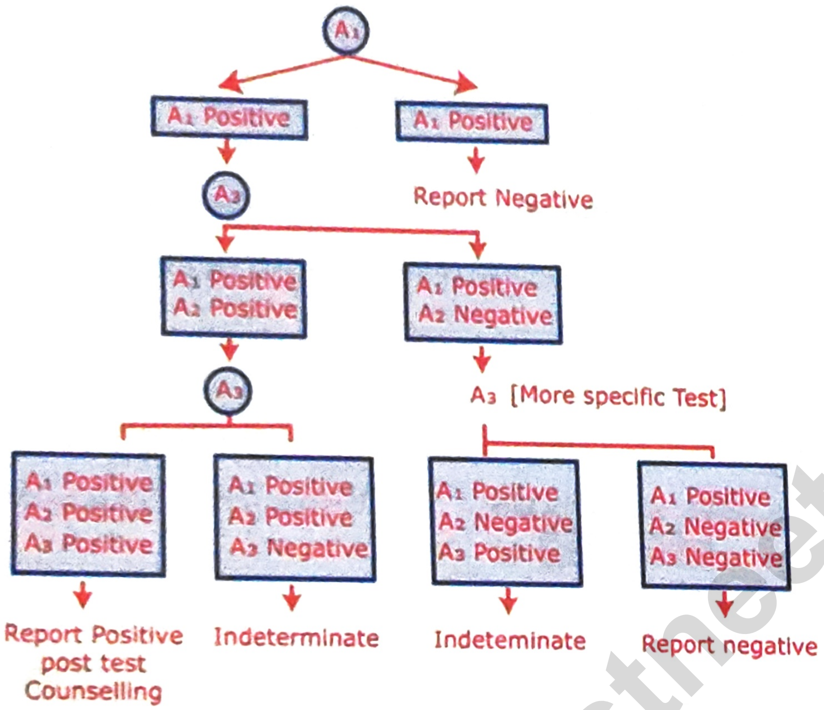
STRATEGY III / ALOGRITHM III

- done to detect HIV infection in Asymptomatic individuals
- 3 test Kits are required



CLINICAL FEATURES

1. ACUTE HIV SYNDROME

- Occurs 3-6 wks after Primary infection
- Burst of plasma viremia +nt
- Typically present as Acute Viral syndrome resembling Acute Infectious mononucleosis [Glandular Fever]
- In most of cases, Acute stage is followed by prolonged period of Clinical latency

2. ASYMPTOMATIC STAGE [CLINICAL LATENCY]

- Median time of latency → 10 yrs
- After Acute HIV syndrome, most of patients develop a clinically asymptomatic stage
- Active viral replication is ongoing & progressive
- Replication occurs mainly in Lymphoid tissues [not in blood]
- It is clinical latency NOT MICROBIOLOGICAL LATENCY
  - Low level of viremia +nt

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- Pt. is infective
- Steady progressive decline [not rapid] of CD_4 T cell count
- Average T cell decline → 50/ml per year
- Rate of disease progression α HIV RNA levels

• Persistent generalized lymphadenopathy
  - 1cm in diameter for › 3 months in 2 or more non-conigous Extra Inguinal sites

• AIDS Related complex
  - Constitutional symptoms or minor opportunistic infections

3. SYMPTOMATIC DISEASE

- Occurs when CD_4 count is very low
- Opportunistic infection & Neoplastic diseases present

CD4 T cell count	Opportunistic infections
400 – 200	Mycobacterium TB Candida (oral Thrush) Herpes zoster Koposi sarcoma Hairy leukoplakia Herpes simplex
Below 200	P →Pneumocystis carinii C →Cryptococcus neoformans → Coccidioidomycosis →Cryposporidiosis T →Toxoplasma gondii
Below 50	MAC [Mycobacterium Avium Intracellulare] CMV retinitis CNS lymphoma

CDC AIDS CASE DEFINITION (with/without HIV serology)

- Pnemonic →6C, 2P & KMH
- If any of following disease is present, it is indicative of AIDS

1) Candida →Esophagus, Trachea, Bronchus

PrepLadder

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2) C. neoformans Meningitis (Extrapulmonary)

3) Cryptosporidium parvum

- Chronic diarrhea (> 1 months)
- Wasting happens

4) CMV Colitis/ Esophagitis

- Serpiginous ulcers are seen
- Serpiginous ulcers also seen in Crohn's disease

5) Cerebral Toxoplasmosis → Seizures present

6) (Primary) CNS Lymphoma

- Cerebral edema present
- Seizures present (BBB damaged)
- Basal ganglia involved (Eccentric dot sign on NCCT head)

*Coccididomycosis, Pulmonary TB, Extra pulmonary TB all these three need positive serology for HIV to mention it as AIDS case

7) P. Jiroveci (CD_4 <200)

8) Progressive multifocal leucoencephlopathy (PML)

- Subcortical white matter lesions seen in Parietal & Temporal lobe
- Caused by JC virus
- Hyperintense lesions on CT (T_2 WI)

9) HSV esophagitis

- Presents with Odynophagia (difficulty in swallowing)
- Solitary large 'Punched Out' ulcer seen
- Mucocutaneous ulcer > 1 month

10) Kaposi sarcoma < 60 yrs

11) M.A.I. (Mycobacterium Avium Intracellulare)

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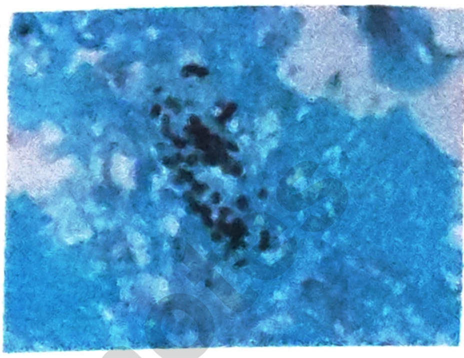
Cd_4 counts below which infections happen

CD4 count	infection
< 500	TB
< 200	P. Jiroveci
< 100	Cerebral Toxoplasmosis
< 50	CMV Retinitis M.A.C.

PrepLadder

P.Jiroveci Pneumonia

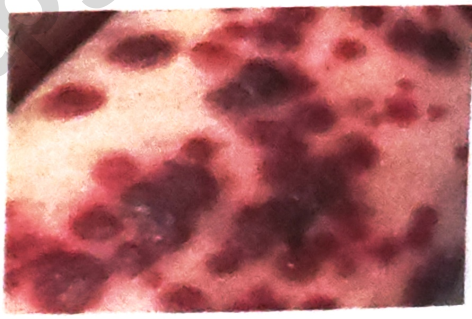
- Perihilar shadows (B/L)
- AIDS positive
- O_2 saturation is less
- BAL, Gastric washings taken → GMS staining



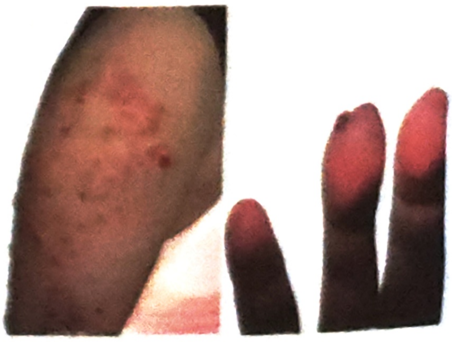
GMS staining of P.Jiroveci

Kaposi sarcoma

- Reddish brown lesions on body which bleed on touch
- D/D → Bacillary Angiomatosis
  - caused by Bartonella henselae



Kaposi sarcoma



Bacillary Angiomatosis

Prophylaxis Guidelines for Opportunistic infections in AIDS

1) P. Jiroveci → Cotrimoxazole
2) Toxoplasma
   - For Prevention of Encephalitis → Cotrimoxazole
   - Toxoplasma with Encephalitis
     - Pyremethamine + Sulphadiazine + Leucovorin
3) M. tb with PPD > 5mm → Latent TB diagnosed
   M. tb with IGRA → Latent TB diagnosed
   - Give (Isoniazid + Pyridoxine) x 9 months
4) M.A.I. (Mycobacterium Avium Intracellulare)
   - Clarithromycin + Ethambutol Rifabutin
5) Varicella Zoster → VZIg
6) Cryptococcus → Fluconozole
7) Histoplasma → Itraconazole
8) Salmonella → Ciprofloxacin
9) Bartonella henselae → Doxycycline
10) CMV → Valcyclovir

Respiratory disease in AIDS patient

- S. pneumoniae > M. tb > P. Jiroveci
- X-ray findings
  - Perihilar opacities → P. Jiroveci pneumonia
  - Consolidation patch → Pneumococcus pneumonia

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PrepLadder

- Multiple opacities / Snow storm appearance/ Hazy appearance →Miliary TB

Ques. Fever with shortness of breath, chest pain, H/O wt. loss CXR → Perihilar opacities seen, LDH ↑ , 1, 3 glucan test positive, DLCO → decreased, Diagnosis ?

Ans. P. Jiroveci pneumonia (PJP)

- 1, 3 glucan test →Positive in Candidiasis, Aspergilosis, P. Jiroveci infection
- Rx of PJP
  - Cotrimoxazole
  - IV Pentamidine (If patient allergic to Cotrimoxazole)
- Steroids are indicated when
  - $PaO_2 < 70$ mmHg
  - A- a gradient > 35 mmHg

M. tb with HIV

- M. tb →More serious
- M. tb cavity seize →smaller →Disseminate more
- TB + HIV coinfection
  - Treat TB first f/b HIV treatment
  - ATT for →2 weeks →cART (combined ART)
  - Treat HIV +ve Patient irrespective of CD_4 count

*Invasive Aspergillosis is not seen in HIV +ve patient

Extra-Pulmonary manifestations of HIV

- Spleen involvement
- Choroid involvement
- Ear canal involvement

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PrepLadder

GIT manifestations of AIDS

- Oropharyngeal Candidiasis
- Odynophagia
- Black hairy tongue →EBV
- Chronic diarrohea (Stool sample)

↓

Modified Zn stain (Kinyoun stain)

↓

Look for

- Cryptosporidium →4-6 µm
- Cyclospora →8-10 µm
- Isospora belli →23-36 µm

CNS manifestations of AIDS

- Soap bubble appearance in any part of brain
  - Diagnosis →Cryptococcal meningitis
  - Rx →(Liposomal Amphotericin B + Flucytosine) x 2 wks
    f/b maintenance on Oral Fluconozale x 8 wks
- Eccentric dot sign in Basal ganglia, Seizures present, IgG Anti Toxoplasma Ab +ve
  - Diagnosis →Cerebral Toxoplasmosis
  - Rx →Pyrimethamine + Sulfadiazine + Leucovorin
- Subcortical Hyperintensity in T_2WI
  - Diagnosis →PML (caused by JC virus)
  - Lab Dx
    - PCR of CSF →JC virus is found
    - No Rx available, Fatal disease
- Space occupying lesion with edema
  - Diagnosis →Primary CNS Lymphoma
  - Lab Dx →Stereotactic Biopsy

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02:34:59

FAMILY	GENUS	Imp. SPECIES
Togaviridae	Alpha virus	Chikungunya Eastern Equine Encephalitis virus Western Equine Encephalitis virus
Flaviviridae	Flavi virus	Japenese Encephalitis B, Dengue West nile Fever Yellow Fever Kyasanur Forest Disease virus Zika virus, Hep C
Bunya viridae	Bunya virus	California encephalitis
	Phlebo virus	Sand Fly fever virus Rift Valley fever virus
	Nairo	Crimean congo Hemorrhagic virus Ganjam virus
	Hanta	Hantan, Seoul, Puumala
Reo viridae	Orbivirus	Colorado Tick Fever
Rhabdo viridae	Vesiculo virus	Chandipura virus Vesicular stomatitis virus

Hantan → Causes Hantan Renal syndrome & Hantan Pulmonary syndrome
→ a/w Rodent faeces

INCUBATION PERIODS

- Dengue → 5-6 Days
- Chikungunya → 5-6 Days
- Japanese encephalitis → 5-15 Days
- Yellow Fever → 3-6 Days
- Kyasanur Forest Disease → 4-8 Days

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JAPANESE ENCEPHALITIS
02:41:40

- Reservoir → Ardeid birds [white cranes]
- Transmitters → Culex tritaeniorhyncus, Culex vishnui
- Amplifiers → Pigs
- Mortality → 30%

CULEX MOSQUITO

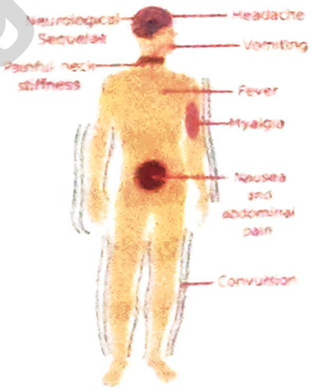
DENGUE
02:44:23

- Serotypes → 1, 2, 3, 4, 5
- Transmitted by → Aedes aegypticus
- ADE [Antibody Dependent Enhancement]

Serotype 1 — infected 1st time & recovered
↓
Serotype 2 — infected 2nd time
↓ Leads to

DHF [Dengue Hemorrhagic Fever]

DSS [Dengue Shock Syndrome]



JAPANESE ENCEPHALITIS

- ANTIBODY DEPENDENT ENHANCEMENT → Instead of protection in 2nd time infection by antibodies, there will be promotion to more severe form of disease
- Maximum chances of landing into DHF seen in
  - Type 1 f/b Type 2 infection

Aedes aegypticus

- DHF
  - Fever – Acute, high & continuous
  - Tourniquet Test +ve
    - > 200 petechial lesions/ Sq. inch seen
  - Thromocytopenia → < 1,00,000
  - Hemoconcentration → Hematocrit ↑ by 20%

- DSS
  - DHS + weak & rapid pulse
  - Narrow pulse pressure
- Break Bone Fever occurs in Dengue

PrepLadder
https://t.me/Latestneetpgnotes

- LAB DIAGNOSIS
  1. NS 1 ANTIGEN TESTING
     - done in the initial 2 days
     - NS → Non Structural or Non Specific protein
     - done by ELISA
     - upto 5th day → IgM capture ELISA can be done
  2. PCR For Dengue virus & its serotypes

WHO case definition for Dengue fever

Acute febrile illness with 2 or more of the following

- Headache
- Retro orbital pain
- Myalgia
- Arthralgia
- Rash
- Hemorrhagic manifestations
- Leucopenia
- Hepatomegaly

DENGUE FEVER RASH

Dengue Hemorrhagic Fever: Pathogenesis

- Secondary infection with another serotype leads to Antibody Mediated Enhancement
- Heterotypic antibodies are non protective & fail to neutralize the virus
- Virus antibody complexes taken up Monocytes
- Virion multiplication in Human Monocytes is promoted
- Activation of CD_4 & CD_8 lymphocytes →Release of Cytokines
- Complement system activated with depression of C_3 & C_5
- Activation of Compliment

↓

Increased Vascular permeability

↓

Loss of Plasma from Vascular compartment

↓

Hemoconcenteration & shock

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PrepLadder

- Disorder of Hemostasis involving Thrombocytopenia, Vascular changes & Co-agulopathy
- Severe DHF with features of shock →DSS

DHF: WHO criteria for Diagnosis

Often occurs with defervescence of fever, swelling

All of the following must be present:

- Fever
- Hemorrhagic tendencies:
  - +ve Tourniquet test
  - Petechiae, Ecchymosis or Purpura
  - Bleeding from other sites
- Thrombocytopenia (1,00,000/cu mm)
- Evidence of Plasma leak
  - Rise in Hematocrit > 20% above average
  - Drop in Hct
  - Pleural effusion/ Ascites/ Hypoproteinemia

DSS: WHO criteria for Dx

All of the above features of DHF + evidence of circulatory failure:

- Rapid, weak pulse
- Narrow pulse pressure 20 mm Hg
- Cold clammy skin
- Restlessness
- Often present with Abdominal pain; mistaken for Acute Abdominal emergency

DF/DHF	Grade	Symptoms	Lab
DF		Fever with 2 or > of: Headache/Retroorbital pain, Myalgia, Arthralgia	Leucopenia, Occasionally Thrombocytopenia, No evidence of plasma leak
DHF	I	Above features + positive Tourniquet test	Platelets < 1,00,000, Hct rise > 20%
DHF	II	Above features + spontaneous bleeding	Platelets < 1,00,000, Hct rise > 20%
DHF	III/DSS	Above features + s/o Circulatory failure	Platelets < 1,00,000, Hct rise > 20%
DHF	IV/DSS	Profound shock with undetectable BP and Pulse	Platelets < 1,00,000, Hct rise > 20%

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*In DHF II → Hct can fall (instead of rising) d/t Spontaneous bleeding

Immune response to Dengue infection

- Primary Infection: IgM antibody in late acute/ convalescent stage; later IgG which lasts for several decades
- Secondary Infection: High IgG level, small rise in IgM
- Cross reactions with other Flaviviruses
- Infection with one serotype does not protect against other serotypes

Management of DHF

- Hospitalise the patient
- Closely monitor for Shock: repeated Hct measurements
- If Hct rising by > 20%, IV fluids as 5% deficit
- Start with DNS 6-7 ml/kg/hr.
- Patient improves → reduce IV fluids gradually over 24-48 hrs
- No improvement → IV fluids upto 15 ml/kg/hr → Colloid solution

DHF: Hct > 20% above normal

↓

Start IVF RL or DNS @ 6-7 ml/kg/hr

Monitor Hct, HR, Pulse pressure, Input-Output

- Improves → Hct ↓ BP ↑
  ↓
  Reduce to 3 ml/kg/hr
  ↓
  Further improvement
  ↓
  Discontinue IVF after 24-48 hrs
- Hct ↑, Pulse pressure ↓, HR ↑
  ↓
  ↑ to 10 ml/kg/hr
  if no improvement → 15 ml/kg/hr
  ↓
  Unstable vitals
  ↓
  CVP line, Urinary catheter, Rapid fluid bolus
  - Hct rises → Colloids
  - Hct falls → Blood Transfusion

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PrepLadder

Criteria for Discharge

Patient with DHF or DSS may be discharged from Hospital when they meet following criteria

1) Afebrile for 24hrs without Antipyretics
2) Good appetite, clinically improved condition
3) Adequate Urine output (1ml/kg/hr)
4) Stable Hematocrit level
5) Atleast 48 hrs since recovery from Shock
6) No Respiratory distress
7) Platelets count > 50,000 cells/ µL

Vaccine of Dengue

- CYD-TDV (Live Attenuated Vaccine)
- 3 doses → 0, 6 & 12 months
- Vaccine is not much effective & has high failure rate
- Can cause ADE (Ab dependent enhancement) so withdrawn in many countries

YELLOW FEVER

03:19:10

- 1st viral disease to be discovered
- Not present in India
  - Prevented by Yellow fever vaccination
  - Vaccination certificate is valid for life time
  - 17 D vaccine is used
- Transmitted by Aedes aegypticus

KFD

03:21:43

- Prevalent in Karnataka
- Mild hemorrhagic Fever
- Reservoirs → Squirrels / Rats
- Transmitter → Tick
- Amplifier → Monkeys

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CHIKUN GUNYA

03:23:18

- Means Bending upon
- Immense Joint pain present → may extend upto 2 yrs after Acute attack of Chikungunya
- Chikungunya → never complicates

TRANSMISSION

AEDES	CULEX
Y → Yellow Fever	B → Bancroftian Filariasis
D → Dengue	E → Encephalitis Japanese
R → Rift Valley Fever	W → West Nile Fever
C → Chikungunya	A → Arthritis like features

Arboviruses → Culture → on now born suckling mice [< 48 hrs]

FILOVIRIDAE

03:27:26

IMPORTANT SPECIES

- MARBURG
  EBOLA VIRUS
- Filamentous in Shape
- MOT → Contact with the secretions of infected patients
- IP → 3-15 Days
- BSL 4 [Bio safety level 4] is maintained
- IOC → PCR For Ebola virus

ARENA VIRIDAE

03:29:29

- Segmented genome is present
- Sandy appearance on Electron microscopy
- Important Viruses
  - Lymphocytic chorio meningitis virus
  - Lassa Virus
  - Junin Virus
  - Machupo virus

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PrepLadder

REO VIRIDAE
03:31:09

- ds RNA Virus
- Have segmented genome
- Important Viruses
  - Ortho reo Virus
  - Orbi virus
  - Rota Virus
  - Colti Virus

ROTA VIRUS
03:32:05

- On Electron microscopy → Wheel with Spoke appearance
- ds RNA Virus
- Segmented genome +nt
- MC cause of diarrhea in children
- IOC → Rota Virus antigen detection in stool by ELISA
- VACCINES → Rotateq/ Rotavarix etc are available
- NSP - 4 → responsible for secretory diarrhea

VIRUS a/w GASTROENTERITIS

- Enteric Adenovirus Type 40, 41 → Infantile diarrhea
- Norwalk Virus
- Calci viruses
- Astro virus
- Rota virus

ONCOGENECITY
03:36:31

DNA VIRUSES

- Papova viridae
- Herpes viridae

Papova viridae

- HPV → Genital tumors

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PrepLadder

Herpes Viridae

- EBV [HHV-4] → Nasopharyngeal Ca
  Burkitt's lymphoma
  Hodgkin/ Non Hodgkin lymphoma
- HHV 8 → Kaposi sarcoma
  Primary Effusion lymphoma
  Castlemann disease
- Hep B → Hepato cellular carcinoma

RNA VIRUSES

- Retro Viridae → HTLV 1 [Adult T cell leukaemia]
- Flavi Viridae (Hep C) → Hepato cellular carcinoma

ZIKA VIRUS

03:40:07

- Family → Flaviviridae
- Transmitted by
  - Aedes
  - Sexual Transmission
  - Mother to child transmission
    - If pregnant mother infected
      ↓
      New born might have Microcephaly, GBS [Guillian Barre Syndrome]

PRIONS

03:41:57

- Proteinaceous infectious particles
- Most resistant forms
- Mechanism of virulence → Abnormal folding [PrP → PrPSC]
- HUMAN DISEASES
  - K → Kuru
  - F → Familial Fatal insomnia
  - C → Creutzfield Jacob disease
  - Gaye → Gerstman Straussler Schienker syndrome

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- ANIMAL DISEASES
  - Scrapie of goat
  - Bovine Spongiform Encephalopathy
  - Mink & Feline Encephalopathy
  - Wasting disease of deer

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PrepLadder

Chapter 13

VIROLOGY & BACTERIOLOGY MUST KNOW UPDATES

EBOLA VIRUS

00:00:28

- discovered in 1976
- Family → Filoriviridae
- elongated Filamentous molecule
- VIRION contains ss, non segmented, negative sense viral genomic RNA
- 5 SUB TYPES
  - ZAIRE EBOLA VIRUS [ZEBOV] → MOST VIRULENT
  - SUDAN EBOLA VIRUS [SEBOV]
  - IVORY COAST EBOLA VIRUS [ICEBOV] → LEAST VIRULENT
  - EBOLA RESTON [REBOV]
  - BUNDI BUGYO EBOLA VIRUS [BEBOV]

EBOLA VIRUS

VIRAL HEMORRHAGIC FEVER caused by

00:02:23

1. FAMILY ARENA VIRIDAE
   - Lassa Fever [Lassa Virus]
   - Lujo virus
   - Argentine [Junin virus]
   - Bolivian [Machupo virus]
   - Brazilian [Sabia virus]
   - Chapare hemorrhagic fever [Chapare virus]
   - Venzuelan [Guanarito virus]
   - White Water Arroyo virus
2. FAMILY BUNYAVIRIDAE
   - Hemorrhagic fever with Renal syndrome [Hanta Virus]
   - Crimean - Congo Hemorrhagic fever virus from genus Nairovirus, reported from Gujrat

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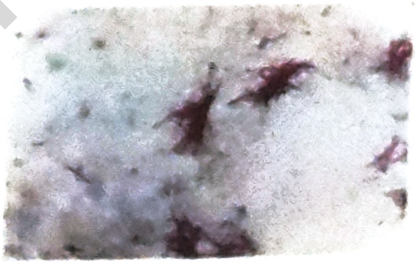
- Garissa virus and Ilesha virus from Orthobunya virus
- Rift Valley fever virus from genus Phlebovirus

3. FAMILY FILOVIRIDAE
   - Ebola virus
   - Marburg virus
4. FAMILY FLAVIVIRIDAE
   - Dengue & Yellow fever and
   - Tick borne encephalitis group
     - OMSK Hemorrhagic fever Virus
     - Kyasanur Forest Disease Virus
5. FAMILY RHABDOVIRIDAE

NOCARDIA

00:06:12

- 1% Acid Fast
- associated with Actinomycetoma
- Pulmonary involvement is + nt mainly
- Culture by Paraffin bait technique



NOCARDIA

ACTINOMYCETES

00:07:38

- Gram positive bacilli with ANTLER-LIKE BRANCHING [Filamentous branching]
- Modified acid fast negative
- Non acid fast mostly
- Mostly non cultured
- Leads to Actinomycetoma
- Causes Oral Actinomycosis (mc location → Cervicofacial)
- Sulphur granules are seen
- Sun Ray appearance

ACTINOMYCETES

CHOLERA TOXIN

00:09:18

- Cholera toxin binds to Enterocytes [Intestinal cells] through interaction of Pentameric B sub unit of toxin with GM_1 ganglioside receptor on Enterocyte

↓

Endocytosis of Toxin

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PrepLadder

- A/B Cholera toxin undergo cleavage of A_1 domain from A_2 domain & A_1 becomes active enzyme
- In the cytoplasm, active A_1 activates the G protein Gs through an ADP Ribosylation reaction that locks G protein in its GTP-bound form & continually stimulate Adenyl cyclase to produce cAMP
- High cAMP levels

  ↓

  Activates CFTR

  [Cystic Fibrosis Transmembrane Conductance Regulator]

  ↓

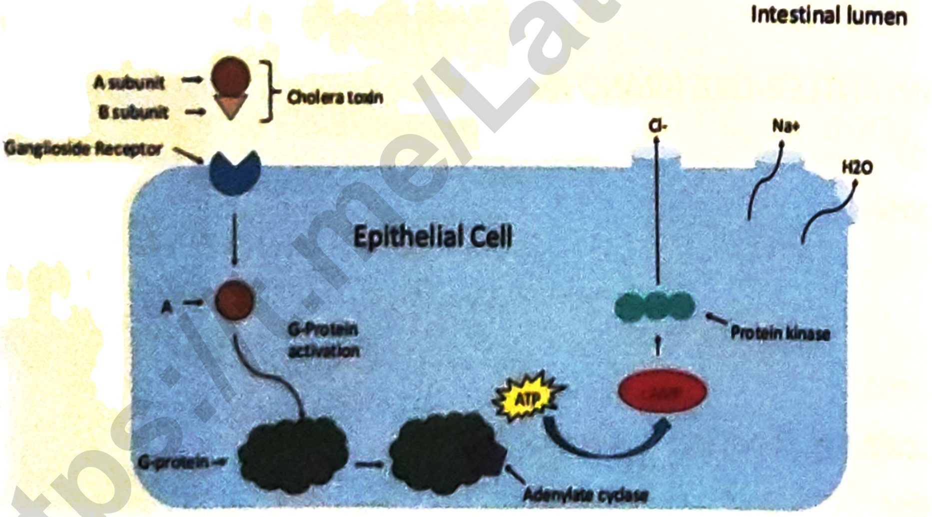
  Dramatic efflux of ions & water from infected Enterocytes

  ↓

  WATERY DIARRHOEA
- ENKEPHALINS in Rx of CHOLERA
  - by direct action on Enterocytes
  - MOA → Binds to Opioid receptors on Enterocytes

    ↓

    Inhibit stimulation of cAMP through G proteins



GAMMA DELTA T CELLS [γδ T cells] 00:15:34

- Have distinctive T cell Receptor
  - most T cells are composed of α & β glycoprotein chains
  - γ δ T cells → composed of one γ & one δ glycoprotein chains
- Less common than αβ T cells but highest in the gut mucosa, within INTRA EPITHELIAL LYMPHOCYTES

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PrepLadder

- Do not require Antigen processing & MHC presentation of Epitopes

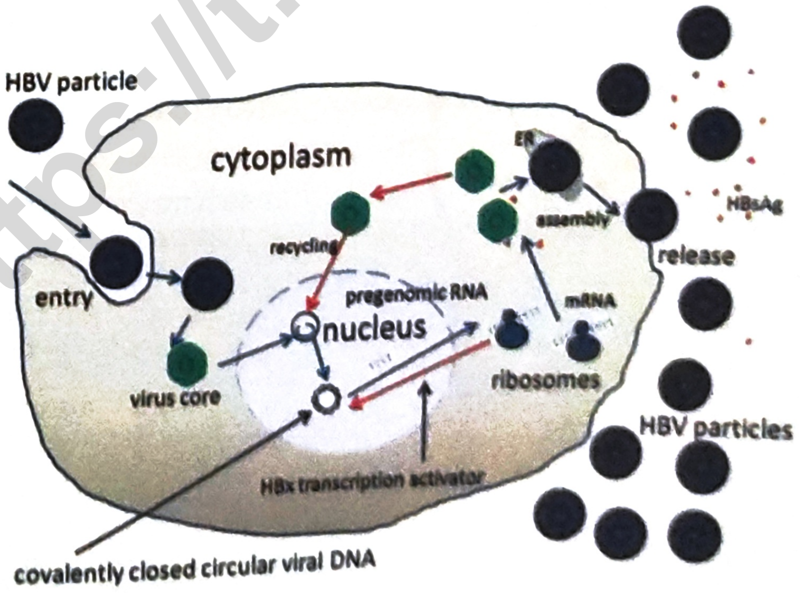
GAS PLASMA STERILIZATION 00:16:54

- GAS PLASMA
  - Fourth state of matter [after solid, liquid, gas]
  - Generate in an enclosed chamber under deep vaccum using Radiofrequency or Microwave energy to produce charged particles from gas.
  - Many of them are FREE RADICALS [Atom with unpaired electron, highly reactive]
  - Gas used is H_2O_2
- Effectiveness depends on
  1. Type of seed gas used
  2. Depth of vaccum

CRYOGLOBULINEMIA 00:20:19

- Characterized by Clonal expansion of Rheumatoid factor expressing B cells in liver, LN & peripheral blood, resulting in presence of Cryoglobulins in circulation
- CRYOGLOBULINS
  - Cold insoluble Immune complexes
  - Contains Rheumatoid Factor, Polyclonal IgG, HCV RNA
  - They precipitate & deposit on vascular endothelium, causing VASCULITIS in Skin, Kidneys & Peripheral Nerves

HEPATITIS B VIRUS REPLICATION 00:21:43



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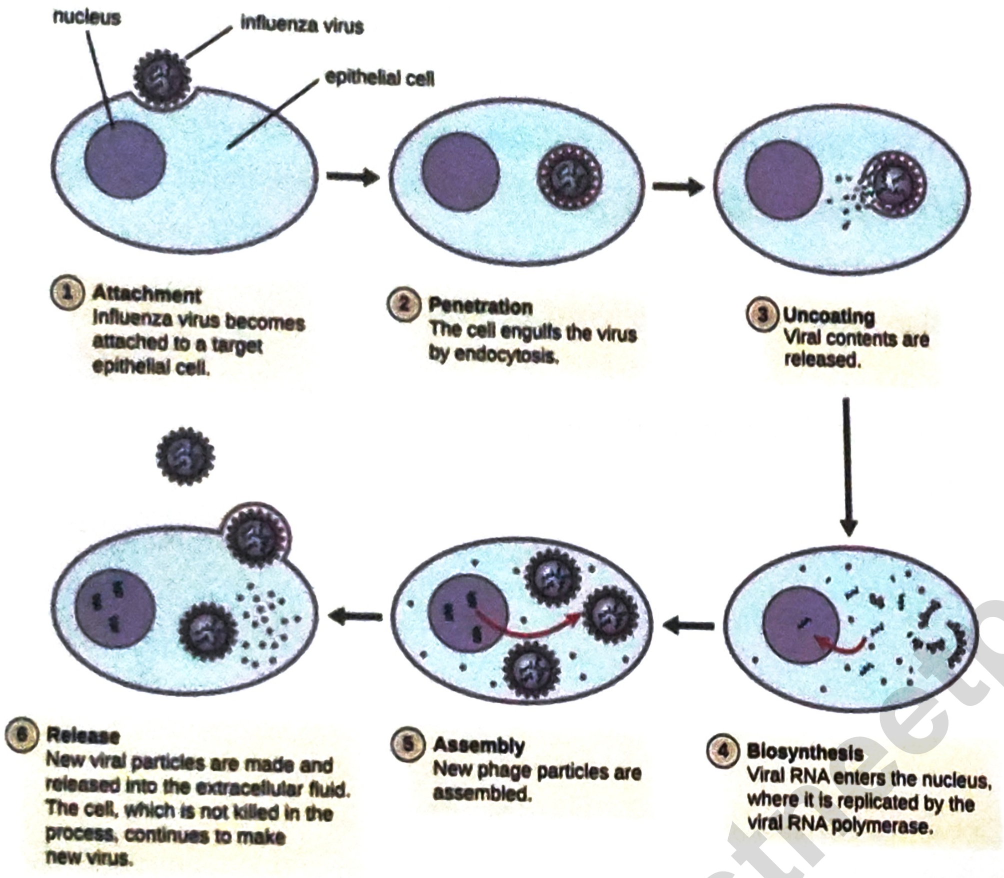
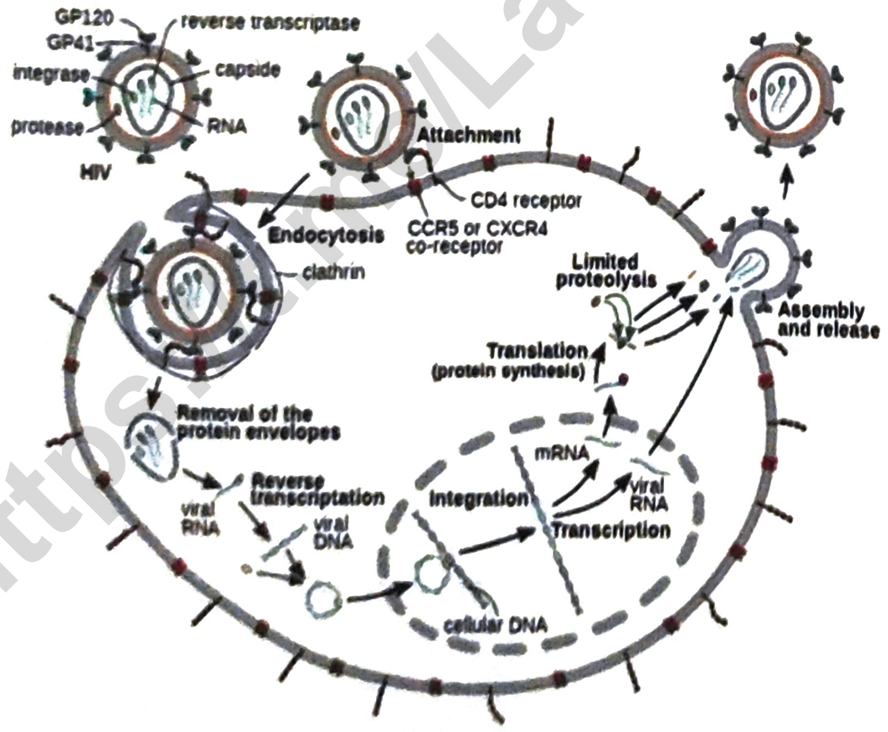
INFLUENZA VIRUS PATHOGENESIS



HIV REPLICATION

00:24:42



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PrepLadder

Chapter 14

COVID-19

- AKA WUHAN Virus
- Caused by SARS COV 2
- Labelled Pandemic on → Feb 11, 2020 by WHO
- Family → Coronaviridae
- Genus → Beta Coronovirus (SARS COV 2)
- Suborder → Cornidovirinae
- Order → Nidovirales
- Realm → Riboviria → Gp. of viruses having RNA dependent RNA Polymerase (RdRP)

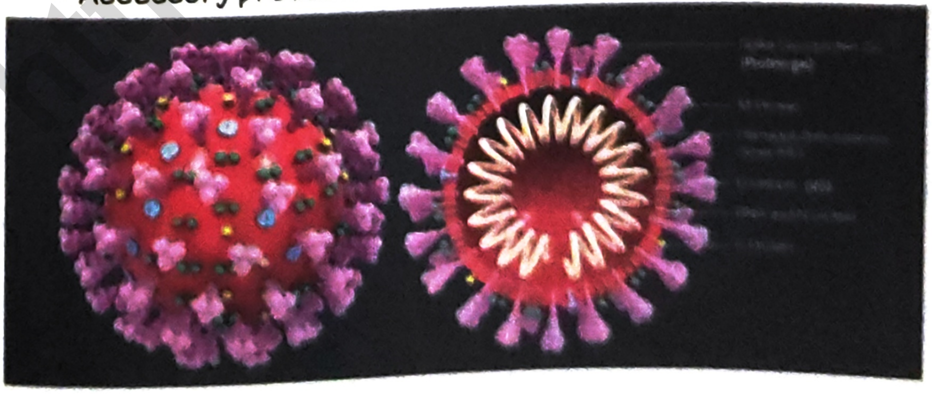
Morphology of SARS COV2

00:02:50

- Virion → Spherical & Enveloped
- Diameter → 100-150 nm
- Most imp. feature → Club shaped Spike projections (coded by S Gene)
- EM (Electron Microscopy) → Crown like appearance (CORONA)

Genome

- Non – segmented, Positive sense ss RNA
- Large genome (30 kbs)
- Contains genes coding for
  - Structural proteins
  - Envelope → coded by E gene
  - Nucleocapsid protein → coded by N gene
  - Membrane Glycoproteins → coded by M gene
  - Accessory proteins



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PrepLadder

Mode of Transmission

00:08:22

- Spreads through infected droplets
- Aerosol spread (more efficient route of transmission)
- Contact transmission
- Spread through
  - Stool contamination
  - Plasma
  - Serum

  } Yet not proved
- Maximum virus shedding → occurs 5-8 hrs before onset of symptoms
- Virus is culturable & can be isolated for next 6-8 days
- Concenterations of SARS COV 2 RNA in upper respiratory specimens have shown decline after onset of symptoms

- For Mild to Moderate COVID-19 patients

  ↓

  RCV (Replication Competent Virus) has not been recovered after 10 days of symptom onset

- For severe COVID-19 infection patients

  ↓

  95% of patient's specimens no longer yield RCV after 14 days

* Patient can be considered non infectious after 2 weeks of symptoms onset

Pathogenesis

00:16:19

- Virus enters upper respiratory tract (URI) & binds to specific receptors
- Initial Asymptomatic phase (1-2 days)

  ↓

  Virus binds to ACE 2 receptors present in human airway with the help of

  ↓

  Receptor binding domain of Spike protein

  ↓

  Leads to Conformational change & fusion of Virus & Host cell membrane

  ↓

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PrepLadder

Following this → Type II Transmembrane Serine protease (TMPRSS2) present on surface of Host cell clears these ACE 2 receptors & further activate the Spike glycoprotein of SARS COV2

↓

Conformational change

↓

Virus entry

↓

Uncoating of Virus
- Release of Nucleic acid in cytoplasm
- Further Replication & Translation in Nuclei

- Upon completion of Machinery → Virus gets assembled in cytoplasm & release by Exocytosis
- Secondary targets → Depend upon ACE 2 receptor numbers
- Max number of ACE II receptor
  - Lungs (Type II Pneumocytes)
  - Gut
  - Heart & kidney

Clinical Features

- 80% of infected person develop mild disease
- 20% of infected person develop moderate to severe disease
  - Out of this 20%,
    - 15% → Respond well to O_2 therapy
    - 5% → May need Ventilatory support

Symptoms in COVID-19

00:26:28

- Dyspnea → Day 5
- Hospitalization → Day 7
- Day 6-9
  - Cytokine storm
  - Release of Interleukins, TNF
  - Mx by Tocilizumab
- Day 9
  - Patient develops ARDS if not managed properly

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Systemic symptoms	Prevalence	Respiratory symptoms	Prevalence	Miscellaneous symptoms	Prevalence
Fever	75-81%	Cough	54-60%	Chest pain and tightness	4-10%
Fatigue	27-35%	Dyspnea	19-28%	Otalgia	1-11%
Myalgia	14-19%	Wheeze	9-26%	Diarrhea	8-12%
Rigors	13-22%	Hemoptysis	1-2%	Nausea	3-10%
		Loss of smell (Hyposmia)	4-55%	Abdominal pain	2-7%
		Sore throat	10-14%	Vomiting	2-8%
		Nasal congestion	3-7%	Headache	10-16%
		Rhinorrhoea	5-12%	Confusion	7-15%
		Pneumonia		Ataxia	Rare
		Respiratory failure		Conjunctivitis	1-4%

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Lab diagnosis

00:35:54

- Gold standard test → Real time (Reverse transcriptase) PCR
- Direct tests
  1) Antigen detection
  2) RTPCR
  3) Gene xpert (CBNAAT)
  4) Tru NAT
- Indirect test
  1) Antibody detection kits
- Supplementary test
  1) CXR
  2) HRCT
  3) Other lab tests → Eg: Look for Interleukin levels in case of Cytokine storm

PrepLadder

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Or click this page to join

RTPCR

- Take 2 swabs
  - 1 Nasopharyngeal swab (NP swab)
  - 1 Oropharyngeal swab (OP swab)

Steps to take NP swab

1) Ask the Pt. to take off the mask to take NP swab
2) Remove swab from packaging
3) Tilt the patient's head back slightly
4) Insert swab along Nasal septum, just above floor of Nasal passage to the Nasopharynx until resistance is felt
5) Gently Rub & Roll the swab, leave swab in place for several seconds to absorb secretions
6) Remove swab slowly while rotating

Steps to take OP swab

1) Insert swab into Posterior pharynx & Tonsillar areas
2) Rub swab over both Tonsillar pillars & Posterior Oro-pharynx

- NP swab & OP swab

↓

Break the swab upto the length taken

↓

Put in VTM

↓

Transport at 2-8°C

- NP swab is better than OP swab

Swabs OF VIRAL SAMPLING

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PrepLadder

VTM (Viral Transport Media)

- Eagle minimum essential medium (commercially available)
- VTM can be easily prepared using

10gm Veal infusion broth

\+

2gm Bovine Albumin fraction V

↓

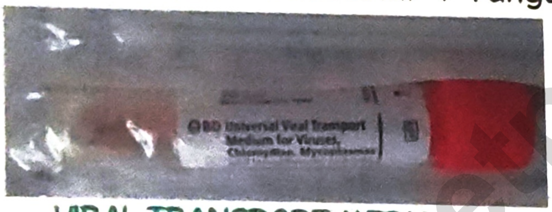
Add into sterile distilled water (400ml)

↓

Add 0.8 ml Gentamicin sulfate solution (50 mg/ml) & 3.2 ml Amphotericin B (250 µg/ml) to avoid Bacterial & Fungal contamination

↓

Sterilize by Filtration



VIRAL TRANSPORT MEDIA

Genes targeted in RTPCR

- Screening genes (Semi confirmatory genes)
  - N → Nucleocapsid
  - E → Envelope
  - S → Spike proteins
- Confirmatory genes
  - ORF 1a → Open reading frames 1a
  - RdRP → RNA dependent RNA Polymerase
- On RTPCR:

E Gene → +ve RdRP Gene → -ve	Not COVID 19
E Gene → +ve RdRP Gene → +ve	COVID 19 +ve
E Gene → +ve ORF 1a Gene → +ve	COVID 19 +ve

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PrepLadder

RTPCR vs TruNAT

FACTORS	RTPCR	TruNAT
Sensitivity	70-80%	70%
Specificity	95%	90-95%
Genes targeted	• Screening genes –N/S/E genes • Confirmatory genes –ORF 1a, RdRP	• Screening genes – E gene • Confirmatory genes – RdRP
Copies detection	Can detect even 10 copies/ml	Detects 486 copies/ml
Turn around time (TAT)	4-6 hrs	60-90 mins
Type of system	Open system	Cartridge based test
Cost	Cost effective than TruNAT	Expensive kits

CYCLE THRESHOLD VALUE (CT VALUE)

- Relative viral RNA estimation
- CT value depends on PCR kits

  CT value $\propto \frac{1}{\text{Viral RNA levels}}$

  - CT value → 16 →suggests high viral RNA
  - CT value →30 →suggest less viral RNA

↓

Patient won't spread disease to others

IMMUNE CHROMATOGRAPHIC TEST (ICT)

- Can estimate Antigen or Antibody
- Various card tests work on Immunochromatographic principle

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PrepLadder

- Procedure
- Take the swab

↓

Put in virus lysis buffer

↓

Take 1-2 drops & put it on card

↓

Check the results

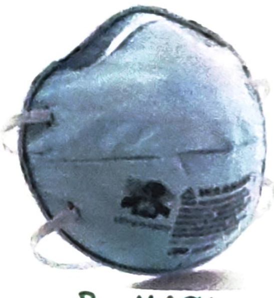
- Use → ICT test
  - Antigen test ↓ Rapid method of diagnosing infection
  - Antibody test ↓ Tells about Seroprevalence in a particular area

N_{95} MASK

Masks

N_{95} is Integral point of PPE

N → Not resistant to oil	R → Resistant to oil	P → oil proof
Types – N_{95}, N_{99}, N_{100}	Types – R_{95}, R_{99}, R_{100}	Types – P_{95}, P_{99}, P_{100}

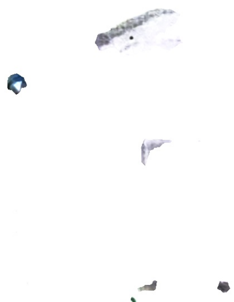


R_{95} MASK

95 indicates → 95% of Airborne particles filtered

99 indicates → 99% of Airborne particles filtered

100 indicates → 100% of Airborne particles filtered



P_{95}

HEPA Filter → High Efficiency Particulate Air Filter

- Filter 99.97% of Airborne particles
- Used in Neurosurgery OT, other surgery OT etc

Basic structure of N_{95} mask

01:03:44

- Outer layer → Spun bond
  - Made up of Non-woven Polypropylene

    ↓

    Blocks droplets & water
- 2nd & 4th layer
  - Made up of Melt blown Non-woven fabric

    ↓

    Protective & Holding layer for 3rd layer (PTFE)

HEPA FILTER MASK

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- 3rd layer
  - Made up of PTFE (Polytetra flouro ethylene)
  - Main filter

Filter efficiency tests

1) Sodium Chloride Aerosol Challenge Test
2) Dioctyl phthalate challenge test

01:08:25

Uses of N_{95}

- Single use (Recommended)

Decontamination & Reuse of N_{95} mask (as per CDC protocol)

1) Vapourised H_2O_2 (Best method)
   - Time of decontamination → 10 mins
   - Mask can be used for 3 cycles

2) UV light (260-285 nm)
   - Time of decontamination → 60 mins
   - Mask can be used for 3 cycles

3) Autoclave
   - Time of decontamination → 121°C x 15 mins
   - Mask can be used for 2-10 cycles

4) Dry heat (Hot air oven)
   - Time of decontamination → 70°C x 1 hr
   - Mask can be used for 2 cycles

5) Air out (Most commonly done)
   - Time of decontamination → 72 hrs
   - Mask can be used for 5 cycles

01:13:26

Drugs used for COVID 19

1) Inhibit Viral RNA Polymerase (RNA synthesis)
   - Includes
     - Remdesivir
     - Favipravir

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PrepLadder

- MOA
  - Adenosine/ Guanosine Nucleotide Analogue
  - RdRP inhibitor

2) Inhibitors of Viral protein synthesis
   - Lopinavir } Protease inhibitors
   - Ritonavir }
3) Viral entry inhibitors
   - Hydroxychloroquine → Increases endosomal pH & inhibits fusion of SARS COV 2 & Host cell membrane
   - Hydroxychloroquine → Also have immunomodulatory action
4) Immunomodulator
   - Ivermectin → Inhibits the Nuclear Symport (Import) of Host & Viral proteins through inhibition of Importin-1 receptors
   - Azithromycin
     - Acts by Induction of Interferon which stimulate genes that attenuate Viral Replication
     - Enhance Neutrophil activation
     - Attenuation of Inflammatory cytokines mainly IL-6 & IL-8
   - Hydroxychloroquine
     - Increases endosomal pH & inhibits fusion of SARS COV 2 & Host cell membrane
     - Hydroxychloroquine → Also have immunomodulatory action
   - Drugs known to prevent/ decrease mortality in COVID 19 patients → Steroids

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Unit: 6 Immunology

Chapter 15

IMMUNOLOGY 1

IMMUNITY → Resistance offered by the host against offenders

00:01:51

INNATE IMMUNITY	ACQUIRED IMMUNITY
Resistance to the infection from birth by its Genetic or constitutional make up	Resistance to infection acquires during life time
Depends on S Species specific I Individual specific R Race specific	Different Forms Natural Active → Infections Natural Passive → mother to child Artificial Active → vaccines Artificial Passive → Immunoglobulins
COMPONENTS - Phagocytes [monocytes, macro phages, neutrophils] - NK (natural Killer) cells - Alternate complement pathway - Acute Phase Proteins [CRP, MBP Serum amyloid protein] - Normal resident Flora - Inflammation, Fever - Skin & mucosal barrier	COMPONENTS - B cell - T cell - Classical complement pathway - Antigen presenting cells [APC]
- Occurs in minute - Diversity Limited - Non specific - No memory	- Occurs in days - Against wide range of infections - Specific - Memory present

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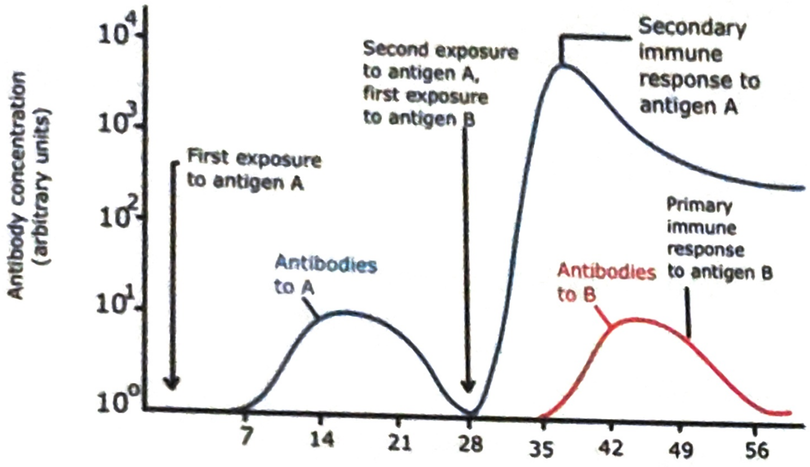
PrepLadder



MBP → mannose Binding Protein

00:10:45

PIR [Primary Immune Response]	SIR [Secondary Immune Response]
→ slow to start Sluggish Short lived	→ Prompt Powerful Prolonged
→ IgM produced mostly	IgM → Ig G Isotype/Class Switching - Occurs d/t change in the constant portion of heavy chain

ANTIGEN

00:18:29

- Any Foreign substance which induces antibody formation under optimum conditions
- HAPTEN → incomplete antigen
  → requires carrier protein to be immunogenic

- FACTORS AFFECTING IMMUNOGENECITY
  1. Nature of the substance
  2. More Molecular weight & size → ↑ Antigenic
  3. Tissue enzyme response
  4. Foreigness α immunogenicity
     α Hypersensitivity

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PrepLadder

NATURE OF SUBSTANCE

Proteins › carbohydrates › Lipids › Nucleic Acids

SUPER ANTIGENS 00:23:41

- non MHC restricted
- attached to TCR on Lateral side on V β (variable β) domain

Induces › 20% T lymphocytes activation

↓

Leads to massive lymphokine release

↓

SHOCK

- Examples

1. STAPHYLOCOCCAL TOXINS
   Enterotoxin A → Food Poisoning
   Exfoliate Toxin A, B → SSSS [Staphylococcal skin scalded syndrome]
   Entero Toxin F → Toxic shock syndrome Toxin

2. streptococcus → scarlet Fever/Erythrogenic toxin A & C
3. Mycoplasma arthritis
4. Malasezzia Furfur
5. Yersinia enterocolitica
6. EBV
7. CMV
8. HIV
9. Rabies virus

T INDEPENDENT ANTIGEN	T DEPENDENT ANTIGEN
- Simple → LPS, capsule, Flagellla - Dose dependent - IgM & IgG3 - No memory - No macrophage processing - Rapidly metabolized - Directly acts on B cells & causes polyclonal B cells activation	- Complex - immunogenic over wide range of dose - All Ab rise - memory +nt - macrophage processing needed - rapidly metabolized

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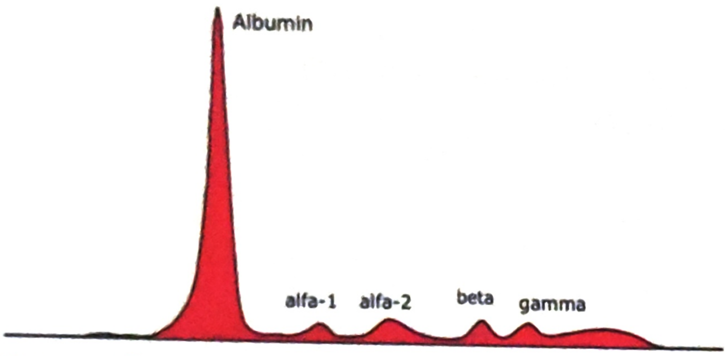
PrepLadder

ANTIBODIES

00:34:01

- Antibodies are immunoglobulins [γ globulins]
- KABAT & TISELIUS found them by doing electrophoresis



Normal Electrophoretic Graph & Blood Proteins

ANTIBODY STRUCTURE

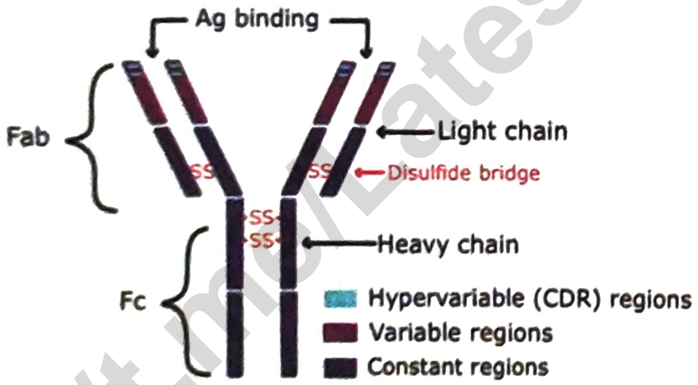
00:35:07

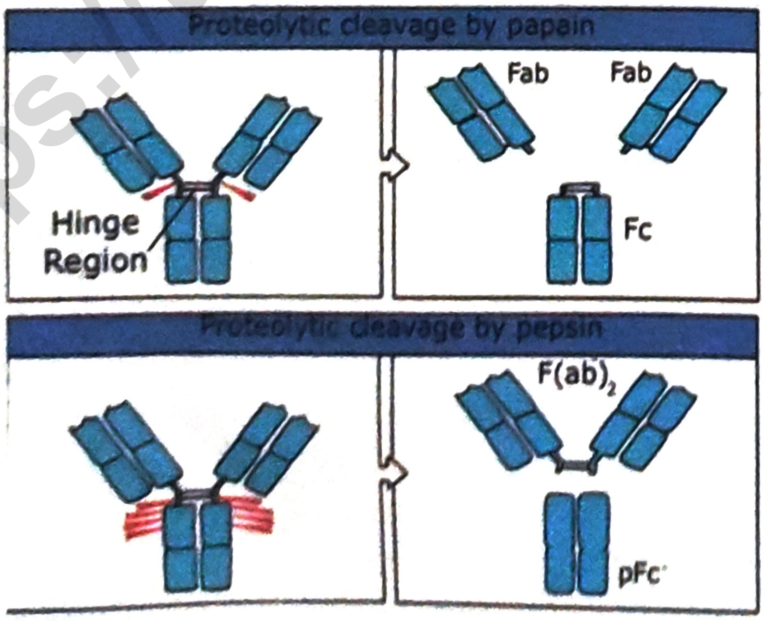
Each antibody consists of 2 Light chains & Each light chain have 1 variable & 1 constant Region

Each heavy chain have 1 variable & ¾ constant regions

Upper portion → Fab [ab = antigen binding]

Lower portion → Fc [c= constant portion]





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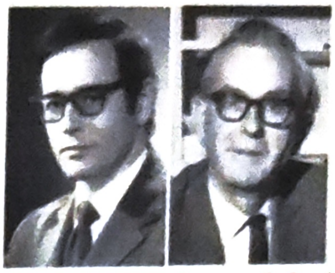
PrepLadder



Gerald M. Edelman Rodney R. Porter

HYPER VARIABLE REGION / PARATOPE/HOTSPOTS

- Actual Antigen binding takes place here
  - Epitope of antigen binds here
- Present in both Light & heavy chains

  Light & Heavy chains binds with each other by Disulfide bond

  Two heavy chains binds with each other by Disulfide bond

  Papain cleaves above the hinge region → 3 Fragments [2Fab + 1 Fc] released

  Pepsin cleaves Below the hinge region → 2 Fragments [F(ab1)2 + Fc1] released

PROPERTIES OF ANTIIBODIES

00:42:34

IgG

- maximum in concentration
- maximum t1/2 → 23 Days
- Sub Groups → Ig G1, G2, G3, G4
- Fc portion of Ig G involves in opsonisation [enhancement of Phagocytosis]
- only Ig which pass through placenta

  Exception → IgG2 doesn't cross placenta
- Ig G4 → doesn't takes place in complement pathway
- IgG3 → doesn't involve in coagglutination
- Role of IgG is more in precipitation [Precipitation > Agglutination]

IgA

- IgA1 → present in serum

  IgA2 → present in mucosal surface & body secretions
- Dimer Form bounded by Joining chains
- mc selective Ig deficiency → IgA

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PrepLadder

IgM
- maximum molecular weight
- aka MILLIONARE MOLECULE
- HAS MAXIMUM SEDEMENTATION CO-efficient
- valency → 2x5 = 10
  Pentamer
- 1st Ig to be produced in any immune response
- MA → Both have Joining chains
- also play a role in Opsonisation

Ig M & D → present over Naïve B cells
IgE → minimum in concentration
- Shortest t1/2 → < 2 Days
- plays a role in HOMOCYTOTROPISM (movement towards most cells)
  - Occurs in Type 1 Hypersensitivity
- only heat labile antibody
- aka REAGINIC ANTIBODY

LIGHT CHAINS 00:54:54
- Kappa : λ → 2:1

ANTIGEN ANTIBODY INTERACTIONS 00:55:49

POST ZONE [Antigen Excess]
ZONE OF EQUIVALENCE [Antigen = Antibody → LATTICE]
PROZONE [Antibody excess]

- Most reactions happen in Zone of Equivalence

SEROLOGICAL REACTION Ag Ab Interactions

AFFINITY → INTENSITY OF ATTRACTION between the antigen & antibody
AVIDITY → STRENGTH OF BOND after the formation of antigen & antibody complex

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PrepLa

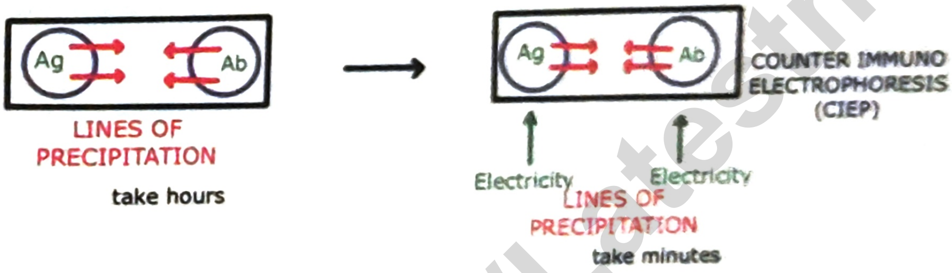
COMPARITIVE EFFICIENCY OF IG IN DIFFERENT SEROLOGICAL REACTION

REACTION	IgG	IgM	IgA
Precipitation	Strong	Weak	Variable
Agglutination	Weak	Strong	Moderate
Complement Fixation	Strong	Weak	Negative
Lysis	Weak	Strong	negative

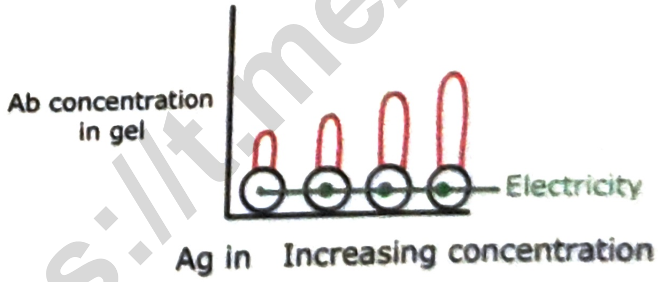
PRECIPITATION

01:01:09

- Soluble antigen + Antibody → Insoluble precipitate
  Eg → ASCOLIS THERMO PPT. TEST → Done For Anthrax antigen
- FLOCCULATION → Modification of precipitation
  Eg → VDRL (Best test to assess prognosis)



- ROCKET ELECTROPHORESIS



IMMUNO DIFFUSIONS

01:07:27

1. Single Diffusion in one Direction [oudin procedure]

Antibody incorporated in agar gel in a test tube

↓

Antigen solution layered over it

↓

Antigen diffuses downward

↓

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PrepLadder

Form a line of precipitation

↓

Number of bands indicates the number of different antigen present

2. DOUBBLE DIFFUSION IN ONE DIMENSION [OAKLAY-FULTHORPE PROCEDURE]

Antibody incorporated in gel

↓

A column of plain agar placed above it

↓

Antigen layered on top of agar

↓

Antigen Antibody move towards each other Through the intervening column of plain agar

↓

Band of precipitate at optimum proportion

3. SINGLE DIFFUSION IN TWO DIMENSIONS [RADIAL IMMUNODIFFUSION]

Antiserum incorporated in agar gel poured on a Flat surface

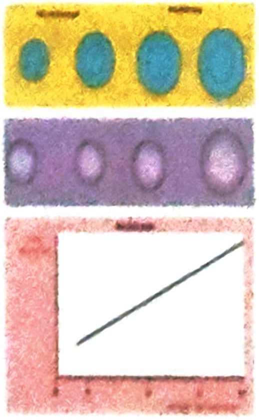
↓

Antigen is added to the wells

↓

Antigen diffuses radially & forms ring shaped bands of precipitation → HALO

Diameter of halo → CONCENTRATION OF ANTIGEN



RADIAL IMMUNODIFFUSION

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PrepLadder

Uses

1. Estimation of Ig in sera
2. Screening of antibodies to influenza viruses

4. Double Diffusion in Two Dimension [Ouchterlony Procedure]

Agar gel poured on a slide

↓

Wells are cut

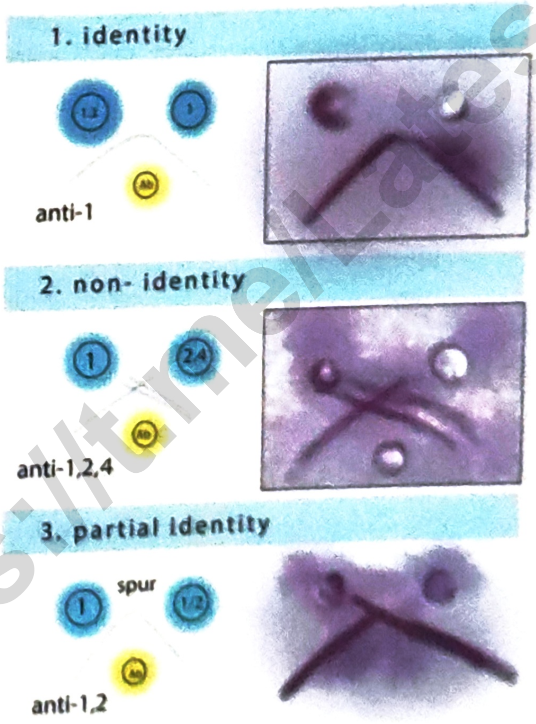
↓

Antiserum placed in the central well
Antigens in the surrounding wells

↓

If two adjacent antigens are
Identical → Line of precipitate Fuse

If unrelated → Line cross each other
Partial identity → spur formation



OUCHTERLONY PROCEDURE

01:21:29

AGGLUTINATION

- Insoluble Antigen + Antibody → CLUMPS [Particulate antigen]
- AGGLUTINATION >>> PRECIPITATION

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PrepLadder

- EXAMPLES

1. WIDAL Test → Antibody against 'O' & 'H' are measured
2. CAT → Cold Agglutination Test → For Mycoplasma
3. SAT → Standard Agglutination Test → For Brucellosis
4. MAT → Microscopic Agglutination Test → For Leptospirosis

5. COOMB'S TEST

DIRECT COOMB TEST → done For Fetal Ab [In vivo test]
INDIRECT COOMB TEST → done For Maternal [In vitro test]

Coomb's sera → Antiglobulin
Incomplete IgG Antibodies

Rh+ Ve Father + Rh - Ve mother → Rh+ ve 1st child
Do incomplete IgG Antibodies testing
If Free incomplete IgG Antibodies in maternal serum
+
O Rh+ RBCs [For tagging incomplete IgG Ab] [In vitro]
+
COOMB SERA
↓
AGGLUTINATION

PASSIVE LATEX AGGLUTINATION TEST

- Precipitation → Agglutination
- Soluble antigen + Latex particle → insoluble antigen
- Examples
  - ASO
  - CRP
  - Rheumatoid Factor Test

NEUTRALISATION REACTIONS

01:33:51

- Toxin + Anti toxin
- Examples
  1. SCHICK TEST → done for Diphtheria
  2. NAGLER'S REACTION → done for clostridium perfringens [α lecithinase]

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PrepLadder

COMPLEMENT FIXATION TEST

01:35:49

1. WASSERMAN REACTION → done For Syphilis
2. SABINFELDMAN DYE TEST → done For Toxoplasmosis

- Antigen + Antibody + RBC system + complement

↓

If Ab present

↓

Ag + Ab complex

↓

Activate complement &
Complement attaches to complex

↓

NO HEMOLYSIS [Ab present]

CARD TESTS

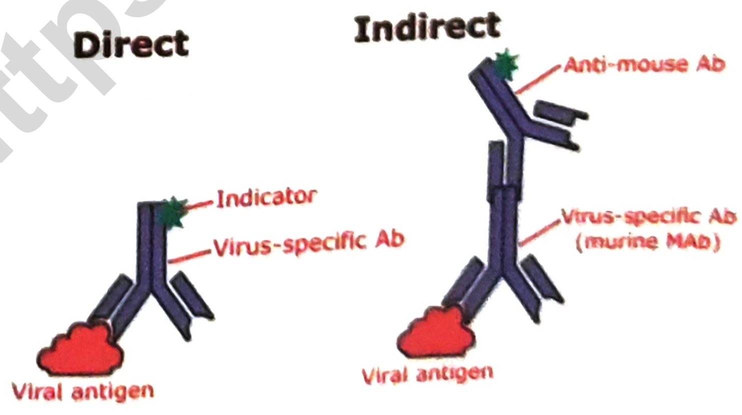
- Based on Immuno chromatographic principle
- Sample runs on nitrocellulose paper
- Eg. HEPA CARD → Test becomes positive if HBs Ag present
- If control is negative, then card test is not valid

Immuno Fluorescence

01:44:00

- Types
  - Direct
  - Indirect

- Fluorescent dyes
  - Fluorescein isothiocyanate
  - Lissamine rhodamine
  - Rhodamine – Auramine etc



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PrepLadder

- DIRECT IMMUNOFLUORESCENCE
  - Detect antigens
  - Principle → Antibodies tagged with fluorescent dyes
    ↓
    Detect unknown antigen
  - Uses

Detection of bacteria, virus, other antigens in blood, CSF, urine faeces, tissues

Diagnosis of Rabies [DFAT- Direct Fluorescent Antigen Testing]

- Disadvantage → Specific Fluorescent labeled Ab required

- INDIRECT IMMUNOFLUORESCENCE → Detection of Antibody

Known antigen
↓
Unknown antibody [serum]
↓
Ab presents – binds with antigen
↓
Fluorescence tagged Ab to human globulin is added
↓
Fluorescence occurs

- Advantage → Single antihuman globulin Fluorescent Ab to any Ag

ELISA [ENZYME LINKED IMMUNO SORBENT ASSAY] 01:49:36

TYPES

1. Direct ELISA
2. Indirect ELISA
3. Competitive ELISA
4. Sandwich ELISA
5. Capture ELISA

COMPONENTS

1. ENZYME CONJUGATE
   - Enzyme conjugated Ab
   - Used as the indicator Ab

- Commonly used enzymes
  a) Horse Radish peroxidase [HRP] - more widely used
  b) Alkaline phosphatase [AP]

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PrepLadder

- There is no significant difference b/w test sensitivities

2. SUBSTRATE & CHROMOGEN

- used as indicator in detecting enzyme – substrate reaction
- commonly used substrates
  a) p- Nitrophenyl phosphate [PNPP]
  b) Hydrogen peroxide – 0.003-0.015%
- Chromogen – are light sensitive so it is necessary to incubate in dark

They are

TMB	→	Tera Methyl Benzidine
ABTS	→	Azino Benthiazoline Sulphonic Acid
OPD	→	O-Phenylene Diamine

- Absorbance (wave length) commonly used is 450nm
  Other wavelength used are 620 nm
  490 nm
  405 nm etc

  They depends upon supplied Kits

DIRECT ELISA → Detects particular Antigen in a sample

- IAg_w + $Ab{*}E_w$ + S → Read

Stage (i)	→	passive absorption of antigen to plate by incubation in defined buffer
Stage (ii)	→	Wash
Stage (iii)	→	Addition & incubation of enzyme labeled antibody
Stage (iv)	→	Wash
Stage (v)	→	addition & incubation of color development system

INDIRECT ELISA

- A well tagged with HIV Ag taken
- Patient serum with Ab1 added to it
- Ab1 —injected to→ Goat —Produce→ Ab2
- If color appears → +ve
- can measure OD values [Optical Density Values]

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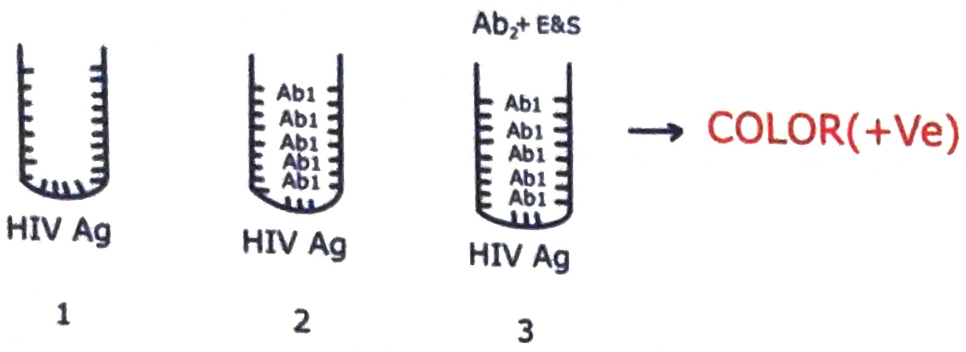
PrepLadder



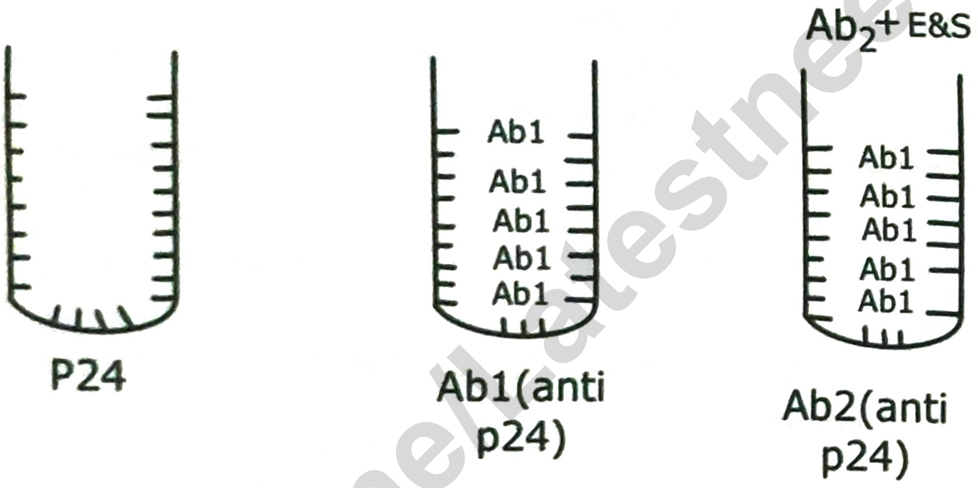
COMPETITIVE ELISA

- A well tagged with P24 Ag taken
- Patient serum with Ab1 (anti P24) added too it

$P_{24} \xrightarrow[\text{injected to}]{} \text{Goat} \xrightarrow[\text{produces}]{} Ab_2\ [\text{anti } P_{24}]$

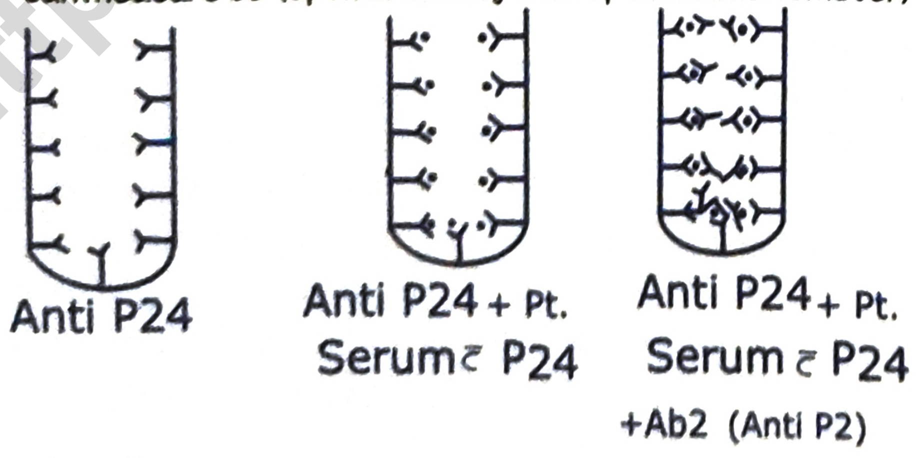
Ab_2 + Enzyme + Substrate added

No Colour → Positive



SANDWICH ELISA

- A well tagged with anti P24 Ab taken
- Patients serum with P24 added to it
- Ab2 [antiP24] + Enzyme + Substrate added
- If colour appears → +ve
- can measure OD (optical density with optical densitometer)



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PrepLadder

HYPER SENSITIVITY

- An altered state of reactivity to an ANTIGEN/HAPTEN in a sensitized host resulting in an unexpected, exaggerated reaction which is injurious to the host leading to tissue damge/ disease/ death

A	→ Anaphylactic hypersensitivity	→	TYPE I
C	→ Cytotoxic hypersensitivity	→	TYPE II
I	→ Immune complex mediated hypersensitivity	→	TYPE III
D	→ Delayed hypersensitivity	→	TYPE IV

TYPE I HYPERSENSITIVITY

02:09:11

- Occurs in 2 phases
  1) Sensitization Phase
  2) Effector Phase

1. Phase Sensitization

- Allergen → Processed by APC &

Peptides of it presented to CD_4 T cells

↓

Activates TH_2 cells

↓

Releases Il_4

↓

Activates B cells

↓

IgE Production

↓

HOMOCYTOTROPISM [movement towards the mast cells]

2. EFFECTOR PHASE

Shocking Dose

↓

Mast cell Degranulation

↓

PRIMARY MEDIATORS

P	→	Protease	
E	→	Eosinophil chemotactic Factor	
N	→	Neutrophil chemotactic Factor	
Hai	→	Histamine Serotonin	} causes vasodilation & Bronchoconstriction

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PrepLadder

SECONDARY MEDIATORS

B → Bradykinins
P → Prostaglandins
L → Leukotrienes
P → PAF [Platelet Activating Factor]
C → Cytokines [Il1, TNF]

LOCALIZED ANAPHYLAXIS / ATOPY

- Run in families
- Eg. Bronchial asthma
  Food allergies
  Allergic rhinitis
  Atopic dermatitis

EXAMPLES

- P-K Reaction
- Theobald smith Phenomenon
- casoni Test

TYPE II HYPERSENSITIVITY

02:22:06

-

  \+ ANTIBODY → complement Activated
- Extracellular matrix antigens
- Exogenous antigens absorbed on host cells
  Eg. Drugs can coat up RBC membrane
- Antigen is bound to cell
- Complement dependent cytolysis
- Complement dependent inflammation
- Complement mediated phagocytosis

} occurs

Complement Mediated Cytolysis → involves MAC (C5-C9)
Complement Dependent Inflammation → involves C3a, C5a (chemo attractants)
Complement Mediated Phagocytosis → involves C3b/C4b (Opsonins)
Best opsonin → C3b

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EXAMPLES

B
Positive → Pernicious anemia
Blood → Blood Transfusion Reaction
Group → Good Posture Syndrome
Graves Disease
IS → ITP
Insulin resistant DM
Mine → Myasthenia gravis

D → Drug induced Hemolytic anemia
A → Auto immune Hemolytic anemia
T → Transfusion Reaction
E → Erythroblastosis Fetalis
Palm → Pemphigus vulgaris

ANTIBODY DEPENDENT CELLULAR CYTOTOXICITY [ADCC]

- Ig G Ab attach to target cell antigens through Fab region
  Fc portion of Ig F inturn binds to Fc receptors of NK cells
- Leads to destruction of the target cell
- used to Killing & destruction of
  Parasites
  Tumor cells
  Graft tissue

ANTIBODY DEPENDENT CELLULAR DYSFUNCTION [ADCD]

- Auto Ab mediated
- TYPES

1. STIMULATORY ADCD
   Eg. Grave's Disease
   - Activation of receptors takes place
   - Auto Ab [Long Acting Thyroid stimulators LATS] produced
   - Stimulate Thyroid to produce hormones

2. INHIBITORY ADCD
   Eg. Myasthenia gravis
   - Inhibition of receptor takes place
   - Anti Ach receptor Ab are produced

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PrepLadder

- Blocking of Ach receptors happens & Leads to Profound muscular weakness

TYPE III HYPERSENSITIVITY

02:45:07

- Immune complex mediated
- Antigen is not bound to cells
- Antigens can be exogenous [bacteria & virus] or endogenous [DNA etc]

Ab Excess

↓

Immune complexes are large & insoluble & tend to localize near the site of antigen administration

↓

Localised Reaction [ARTHUS REACTION]

Localised area of neurosis d/t vasculitis

Ag Excess [monovalent Antigens]

↓

Small soluble complexes tend to travel through blood to various sites & get deposited

↓

Generalised Reaction [SERUM SICKNESS]

↓

Occurs following serum therapy [mostly with Horse antitetanus serum] Manifest after 7-8 Days

Edema, Erythema, Glomerulonephritis can occur

- EXAMPLES

H → Hypersensitivity pneumonitis, HSP

A → Arthus Reaction

R → Reactive arthritis

P → PAN, PSGN

E → Epstein Barr Infection

R → Rheumatoid Arthritis

S → Serum sickness, SLE

TYPE II Lepra Reaction

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TYPE IV HYPER SENSITIVITY

02:56:40

- Delayed Hypersensitivity
- SENSITIZATION PHASE

APC present peptide fragment with MHC_2 to TH_1 cells

↓

TH_2 cells differentiate to from T delayed Hypersensitivity [T_{DTH}] cells

Other T cells are also involved → also act as DTH cells

CD_8 T cells

CD_4 TH_{17} cells

- EFFECTOR PHASE

T DTH cells on subsequent contact antigen

↓

Release cytokines

↓

Attract & recruit inflammatory cells at the sites

CYTOKINES

I → Interferone [most important]

I → Il_2

M → MCAF [Monocyte chemo Attractant Factor]

To → TNF [TUMOR Necrosis Factor]

Medical → MIF [macrophage Inhibition Factor]

Institute → Il_3

Gujarat → GMCSF [Granulocyte Monocyte Colony Stimulating Factor]

IFN → activates resting Macrophages

IL_2 → acts in autocrine manner & helps in proliferation of T_{DTH} cells

PATHOLOGY

- Continuous TDTH reaction For Killing intracellular microbes

↓

Formation of GRANULOMA [TB, Leprosy etc]

T helper cell infiltrate is replaced by macrophages in 2-3 weeks

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Macrophage transforms into

1 Large, flat; eosinophilic cell [Epitheloid cell]
2 Epitheloid cells fuse [with the help of INF] to Form Multinucleated gaint cells

EXAMPLES

TUBERCULIN TEST → Delayed type hypersensitivity

CONTACT DERMATITIS

Nickel, oak poison act as Haptens

↓

Haptens + Skin proteins

↓

Internalized by skin APC [Langerhan cells]

↓

Presented to TDTH cells

↓

Activates macrophages

↓

Release lytic enzymes

↓

Causes skin redness & postule formation

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Chapter 16

IMMUNOLOGY 2

HYBRIDOMA TECHNOLOGY

00:00:15

- Done for mass production of Monoclonal antibodies
- Principle → B cells fused up with Myeloma cells on HAT media [Hypoxanthine Aminopterine Thymine media]
  - Mouse is injected with desired Antigen

    ↓

    Produces B cells [HGPRT +ve]

    ↓

    B cells [HGPRT +ve] + Myeloma cells [HGPRT-ve] on HAT media

- ○ Unfused B cells [HGPRT +ve] → Do not survive
- ○ Unfused Myeloma cells [HGPRT -ve] → Do not survive
- ○ Fused cells [Hybrid cells] → Survive

* Fused cells (Hybrid cells) → B cells + Myeloma cells

- Aminopterine
  - Blocks denovo pathway for Purine synthesis
  - Now every cell will go through salvage pathway to synthesize Purine
- Hybrid cells → Mice → Desired Monoclonal antibodies

COMPLEMENT SYSTEM

00:09:18

- Non Specific Proteins
- Constitute 5% of serum proteins
- Heat labile (56°C x 30 min)
- can blind to Fc portion of Ig

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PrepLadder

SITES OF SYNTHESIS

Liver → C_3, C_6, C_9

GIT → C_1

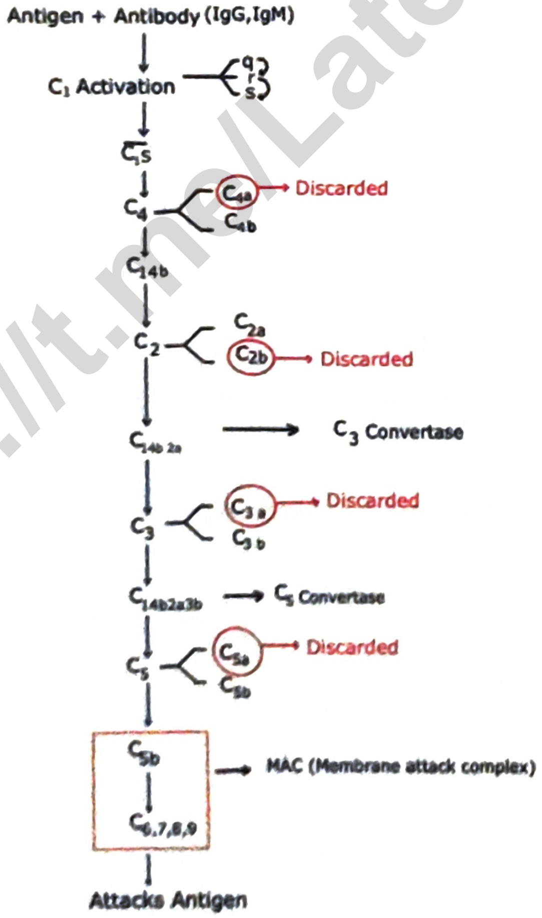
Macrophage → C_2, C_4

Spleen → C_5, C_8

TYPES

	CLASSICAL	ALTERNATIVE	LECTIN
Activators	Ag + Ab complex	Zymogen Endotoxin Nephritic Factor Properidin	Mannose binding Lectin
1st Activated	C_1	C_3	C_4
Part of	Acquired immunity	Innate immunity	Innate immunity

CLASSICAL COMPLEMENT PATHWAY



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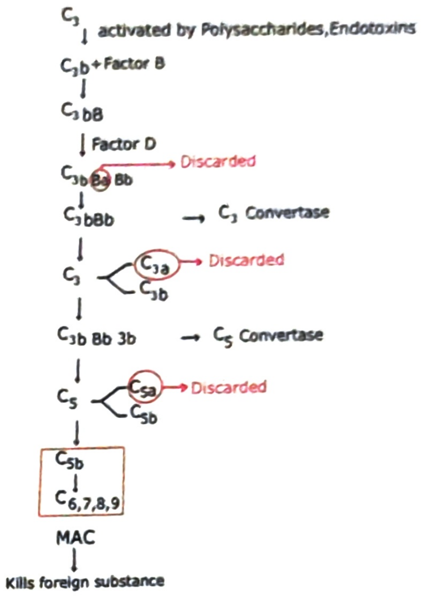
ALTERNATE COMPLEMENT PATHWAY



C_1, C_4, C_2 → will not be consumed in Alternate complement pathway

LECTIN PATHWAY

- Similar to Classical complement pathway, but starts with C_4
- C_1 is not consumed

BIOLOGICAL ROLE OF COMPLEMENT SYSTEM

1) Lysis of target cells
2) Chemotactic & Anaphylactic role → C_{3a}, C_{5a}
3) C_{3b} → best opsonin
4) Mediate Type 2 & Type 3 Hypersensitivity Reactions
5) Auto immune diseases
6) Endotoxic shock

DEFICIENCY/ABSENCE

00:26:33

- MAC Deficiency → Leads to Recurrent Neisserial infections
- Factor D & I absence → Leads to Pyogenic infections
- Factor H absence → leads to Hemolytic Uremic syndrome
- C_1 esterase inhibitor deficiency → Heriditary Angioneurotic Edema (HAE)

– Side effect of ACE Inhibitors → HAE (d/t C_1 esterase deficiency)

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PrepLadder

INTERFERONS

00:29:15

- Induced by virus
- Can confer protection against same virus or unrelated virus
- Not virus specific but species specific
- MOA → inhibit viral protein synthesis
- Secreted by

  L → Leucocytes → IFN α

  F → Fibroblasts → IFN β

  T → T-lymphocytes → IFN γ

- USES
  1. Rx of Hepatitis B & C
  2. Rx of Multiple sclerosis

STRUCTURE OF IMMUNE SYSTEM

00:33:15

MYELOID SYSTEM	LYMPHOID SYSTEM
RBC	Lymphoid cells
WBC	LYMPHOID organs
Platelets	Thymus
Granulocytes	Bone marrow

LYMPHOID ORGANS

00:34:36

Central → Thymus
Bone marrow

Peripheral → Spleen
Lymph nodes
MALT [Mucosa Associated Lymphoid Tissue]
GALT [Gastric Associated Lymphoid Tissue]
Liver

LYMPHOID CELLS

T cells
B cells
NK cells

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PrepLadder

THYMUS

- Develops From 3rd & 4th Pharyngeal pouches
- Maturation of T cells occurs in Thymus

BONE MARROW

- B & T cells originates From BM
- B cells mature in BM

PERIPHERAL LYMPHOID ORGANS

- T dependent area in a LN → Para cortical area
- B dependent area in a LN → Cortical follicle

SPLEEN

- T dependent area in Spleen → around Periarteriolar lymphoid sheath
- B dependent area in Spleen → Germinal follicle [Marginal Zone]

CD MARKERS

T cells → CD_1 to Cd_8

B cells → CD_{10}, CD_{19} to Cd_{23}

B LYMPHOCYTES

00:41:58

- Constitute 10-20% of total lymphocytes
- Also acts as APC [Antigen presenting cells] [with Surface IgM]
- CD_{21} → EBV receptor for B cells
- CD_2 → Measles receptor for B cells
- On Electron Microscopy → shows Microvilli on surface

- Blast Transformers of B cells → Endotoxins (mainly), EBV Ag
- Blast Transformers of T cells → Concavalin, Phytohaemagglutinin

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PrepLadder

- ROSETTES TYPES ON RBC

    B cells → EAC Rosettes

    T cells → E Rosettes

T CELLS

00:46:41

- Constitute 60-70% of total lymphocytes

TYPES

1) CD_4 T CELLS

TH_1 → secretes IL-2, IFN γ

TH_2 → secretes IL-4, 5, 6, 10

- The earliest identifiable T cell precursor in bone marrow are (CD_{34} + Pro T cells)

2) CD_8 T CELLS [CYTOTOXIC T CELLS] → causes lysis of target cells

NK [NATURAL KILLER] CELLS

00:50:23

- Constitutes 5-10% of all lymphocytes
- Kills Virus infected cells & Tumor cells

APCs [Antigen Presenting cells]

↓

Secretes IL-1

↓

Stimulates TH_1 cells

↓

Secretes IL-2

↓

Stimulates NK cells → LAK CELLS [Lymphokine Activated Killer cells]

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PrepLadder

MHC [Major Histocompatibility Complex]   00:53:21

- Present on chromosome 6p

CLASS I MHC GENES

- Present in all nucleated cells
- Provides peptide fragments of cell to CD_8 T cells
- Helps in processing of Virus infected cells & Tumor cells

CLASS II MHC GENES

- DP/DQ/DR
- Glycoprotein present on APC
- Provides exogenous peptide fragments to CD_4 T cells
- Play central role in Transplantation antigens immune response

CLASS III MHC GENES

- a/w Complements → C_2, C_4, Properidin factors, C_3 convertase, TNF, HSP (Heat shock protein)

MHC RESTRICTION

- Only above process will occur

Exogenous antigen
↓
Peptide fragments with MHC II
↓
CD_4 T cells

Endogenous antigen
↓
Peptide fragments with MHC I
↓
CD_8 T cell

MACROPHAGES   01:01:11

- Play a role in Phagocytosis
- Act as APCs

- MODIFIED MACROPHAGES

Peripheral Blood	→	Monocytes
Liver	→	Kupffer cells
Brain	→	Microglial cells
Kidney	→	Mesangial cells
Lungs	→	Alveolar macrophages
Skin	→	Langerhan cells

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PrepLadder

- Acts/ Kills by
  - Phagocytosis
  - Phagolysosomal fusion
  - Lysosomal degranulation
  - O_2 free radical kiling

DENDRITIC CELLS

- APCs
- Derived from BM
- Transported to lymph nodes

INTERLEUKINS

01:05:41

	SECRETED BY	FUNCTION
IL 1	Monocytes Macrophages Dendritic cells	Target vasculature Induces inflammation, fever
IL 2	TH_1 Cells	NK cells → LAK cells
IL 3 [GMCSF]	TH Cells	Granulocyte Monocyte Colony Stimulating Factor
IL 4	TH_2 cells	Class switching to IgE
IL 5	TH_2 cells	Class switching to IgA Eosinophil attractant
IL 6, 10, 13	Same function as IL 4	
IL 8	Macrophages	Neutrophil chemoattractant
IL 12	Macrophages	Promotes TH_1 induction & inhibit TH_2

CHEMOKINES

01:13:33

CxC CHEMOKINES

- Act on Neutrophils
- IL8, IL1, TNF

CC CHEMOKINES

- Monocyte chemoattractant protein 1
- Eotaxin; RANTES

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PrepLadder

C CHEMOKINES

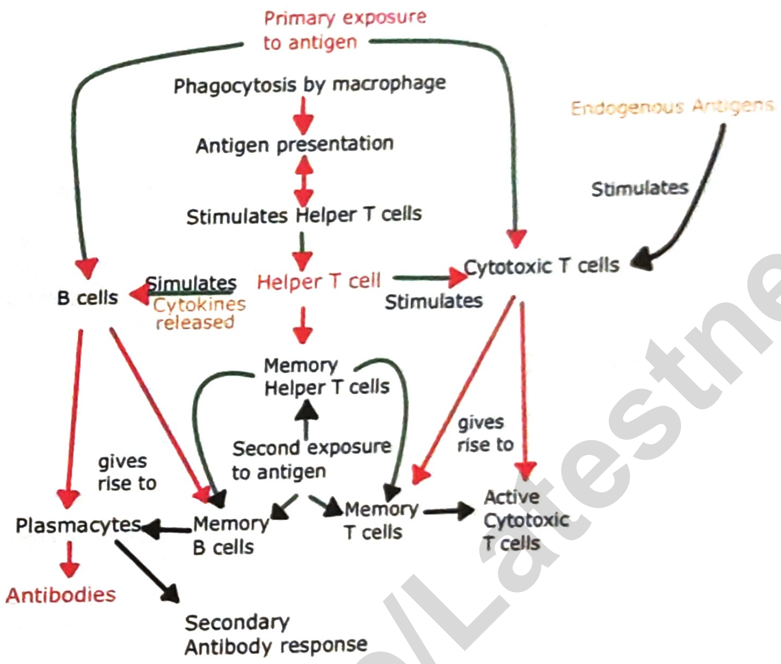
- Specific For lymphocytes & lymphotactin

Cx_3C CHEMOKINES → [FRUCTALKINE]

FUNCTIONS

T cells → Cell mediated Immunity

B cells → Humoral Immunity



ROLE OF CELL MEDIATED IMMUNITY

01:21:25

1) Immunity against intracellular organisms [TB, Leprosy]
2) Immunological Surveillance & immunity against cancer
3) Transplantation immunity
4) Delayed type hypersensitivity
5) Auto immune diseases

ADJUVANT

01:23:44

- ↑ ses the immunogenicity of Ag or Vaccine
- Examples
  - $Al(OH)_3$
  - $AL Po_4$
  - LPS of B. pertussis [in DPT vaccine]

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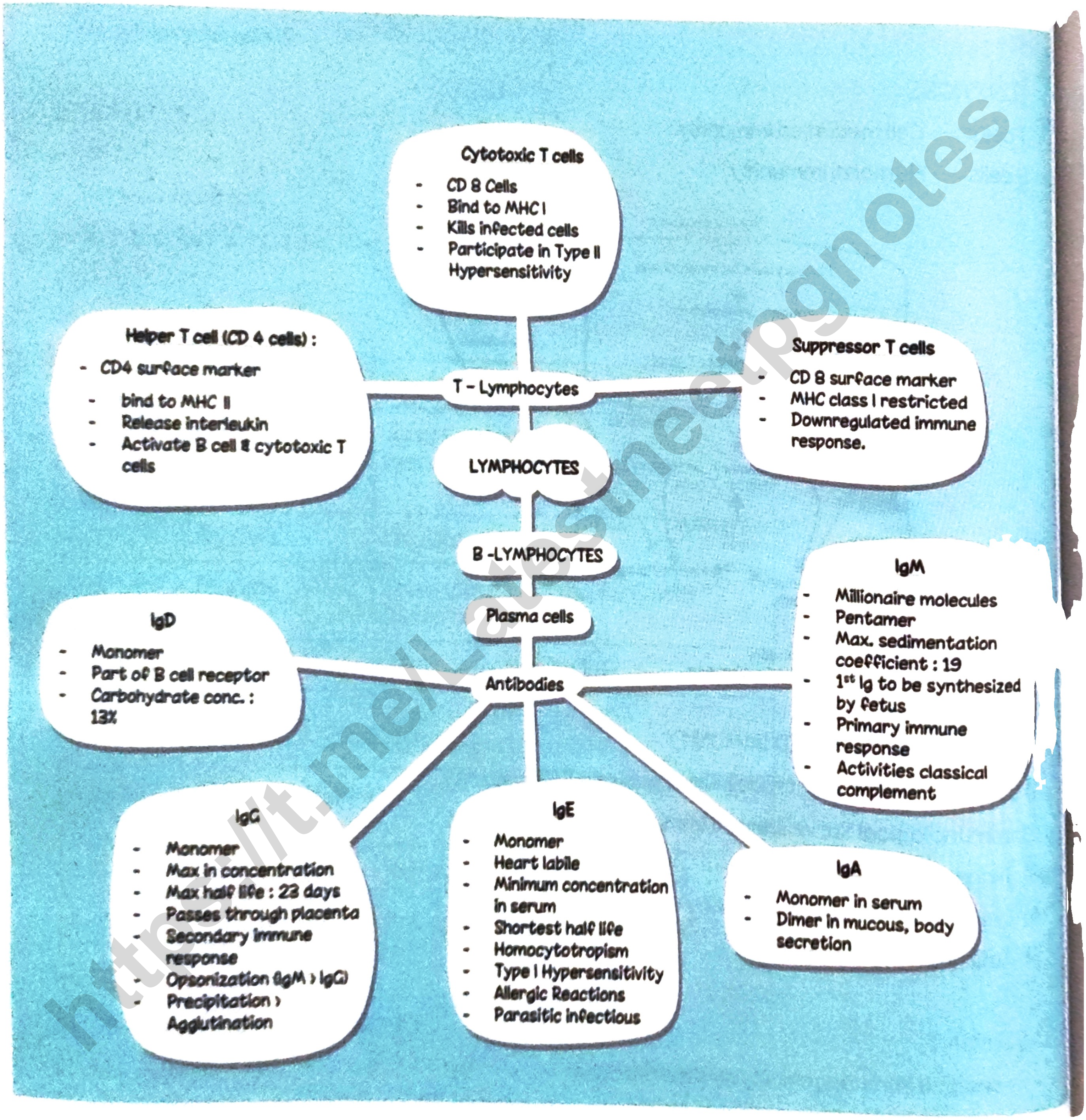
PrepLadder



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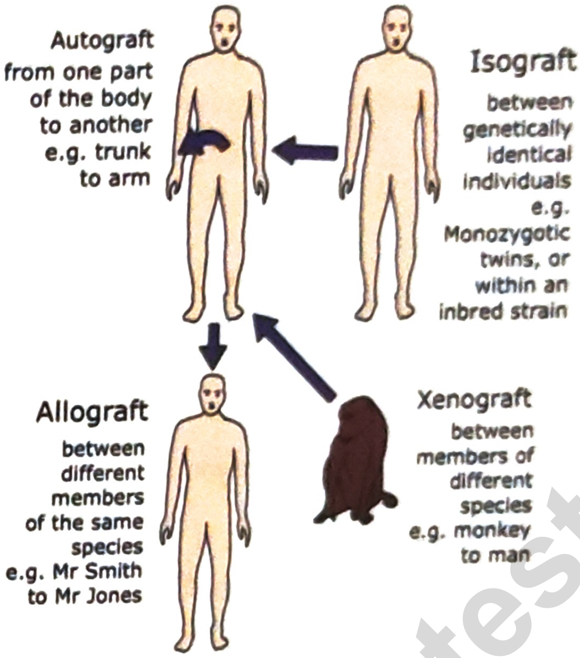
Chapter 17

IMMUNOLOGY 3

GRAFT

00:00:16



XENOGRAFT / ALLOGRAFT REACTION

00:02:28

SERIES OF EVENTS

- Graft is vascularised, healthy in first 3 days
- From 4th day
  - There is inflammation with Lymphocytes, Macrophages

↓

Blood vessels in Graft are occluded by thrombi

↓

Vascularity diminishes & graft undergoes Ischemic necrosis by 10th day

↓

Necrosis & scab like appearance

↓

Sloughs Off [1st SET RESPONSE]

PrepLadder

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- If another graft from the same donor, it will be rejected in an accelerated fashion

↓

Vascularisation commences

↓

Soon interrupted by inflammatory response

↓

Necrosis sets early

↓

Graft slough Off by 6th day

GRAFT REJECTION

00:06:58

TYPE	TIME TAKEN	MECHANISM
HYPER ACUTE	– Minutes to hours	– Preformed Ab
ACUTE	– Days to weeks	– CD_4 T cell activation
CHRONIC	– Months to years	– Fibrosis & Wide spread Arteriopathy [T cell]

GRAFT VS HOST REACTION

BILLING HAM's CRITERIA

1) Graft contains immune competent cells
2) Host is immunosuppressed
3) MHC incompatible [MHC typing not done properly]

- Skin, Liver & GIT are the main targets of GVHD
- GVH in animal model → RUNT DISEASE
    - Cachexia
    - Animal dies

TYPING METHODS

00:14:00

HLA TYPING

1) SEROLOGICAL METHODS
    - Micro cytotoxicity
    - Mixed Leucocyte Reactions

2) DNA PCR TYPING [Best]

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PrepLadder

AUTOIMMUNITY

00:15:35

- Immune response to its own antigens leads to various pathological conditions/diseases

MECHANISMS OF AUTOIMMUNITY

1) Exposure of sequestrated antigen
2) Formation of Neo antigens
3) Immunological damage by cross reactivity
4) Breakdown of immunological homeostasis
5) Exposure of forbidden clones

SEQUESTRATED ANTIGEN

- Lens protein
- Sperm protein

} considered as foreign

FORMATION OF NEO ANTIGENS

- Biological exposure
- Chemical exposure

} Neo antigens

IMMUNOLOGIC REACTION BY CROSS REACTIVITY

- Carbohydrate 'C' cross reacts with Cardiac valves
- 'M' Protein cross reacts with Myocardium

BREAKDOWN OF IMMUNOLOGICAL HOMEOSTASIS

- No coordination b/w Cell mediated immunity & Humoral immunity

FORBIDDEN CLONES are later exposed in young age

EXAMPLES

1) AIHA [Auto Immune Hemolytic Anemia]
2) Myasthenia Gravis
3) SLE [Cause of Death is due to Kidney lesions mainly]
4) Rheumatoid Arthritis
   - IgM against Fc portion of IgG
   - IgM + IgG complex deposited in synovium of Joints & activate complement
5) Hashimoto's Thyroiditis
   - Age group → Middle aged women
   - Infiltration of gland by sensitized lymphocytes leads to

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PrepLadder

progressive destruction of Thyroid follicles

- Presents with Goiter

MECHANISM OF AUTO IMMUNITY IN A NUTSHELL

1) Exposure of sequestrated antigens
2) Formation of Neo antigens
3) Cross reacting antigens
4) Polyclonal activation of B cell
5) Inappropriate expression of MHC class II molecules

IMMUNO DEFICIENCY DISORDERS

00:31:01

DISORDERS OF PHAGOCYTOSIS

1) CGD [Chronic Granulomatous Disease]
   - Deficiency of NADPH Oxidase
   - Leads to recurrent catalase positive infections [Staph & Coliforms]
   - NITRO BLUE TETROLIUM TEST → -ve

2) LAD SYNDROME
   - Absence of Cd_{18}
   - Problem in common β chains of Integrins
   - leads to recurrent infection

3) CHEDIAK HIGASHI SYNDROME
   - Defective Phagolysosomal fusion
   - ↓↓↓ in Melanocytes

4) JOB SYNDROME/ HYPER IgE SYNDROME
   - Deficiency of Th_{17}
   - Leads to recurrent Staph infections, Cold abscess
   - ↑ IgE

5) MPO DEFICIENCY
   - Myeloperoxidase deficiency
   - Mild or none presentation

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PrepLadder

B CELL DISORDERS

00:37:10

1) BRUTON'S AGAMMAGLOBULINEMIA/ X LINKED AGAMMA GLOBULINEMIA

- Defect in B Tyrosine Kinase gene (BTK gene)
- PRO B CELLS —✗→ PRE B CELLS (BTK gene defect)
- Upto 6 months of neonatal life, nothing happens to male child [d/t Maternal Antibodies]; After that severe infections occur

2) X LINKED HYPER IgM SYNDROME

- Deficiency of CD_{40L} on activated T cells
- ↑ Serum IgM
- T cells are normal

3) SELECTIVE IgA DEFICIENCY

- Leads to repeated sino pulmonary infections

4) COMMON VARIABLE IMMUNO DEFICIENCY

- Unknown defect
- B —✗→ CELLS
- Plasma cells
- ↓↓ Immunoglobulins
- Late onset
- Commonly a/w Giardiasis

5) TRANSIENT HYPOGAMMA GLOBULINEMIA

- Delayed onset of normal IgG synthesis
- Can resolve itself upto 2-6 yrs

COMBINED B & T CELL DISORDERS

00:44:27

1) WISKOTT ALDRICH SYNDROME

- WASP gene defect
- R → Recurrent infections
  E → Eczema
  T → Thrombocytopenia

- IgM → ↓ ed
  IgG & IgA → Normal
  IgE → ↑ ed

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PrepLadder

2) ATAXIA TELANGIECTASIA

- defect in Kinase involved in cell cycle
- deficiency of IgA & IgE
- leads to
  - Cerebral ataxia
  - Telangiectasia
  - Ovarian dysgenesis

3) SEVERE COMBINED IMMUNO DEFICIENCY [SCID]

ETIOLOGY

- Defect in common gamma chain of IL-2 receptor
- Defect in receptors of IL4,7,9
- 2nd MC defect → Adenosine deaminase (ADA) deficiency
- Rag 1 & Rag 2 genes defect → Total absence of T & B cells
- Deficiency of PNP [Purine Nucleoside Phosphorylase]

TREATMENT → Bone marrow transplantation

T CELL DEFECTS

00:50:38

1) DI GEORGE SYNDROME

- Chromosome 22q11 defect
- Absent Thymus & Parathyroid glands
  - Presents with
    - Hypocalcemic Tetany
    - Congenital Heart Defects

2) CHRONIC MUCOCUTANEOUS CANDIDIASIS

- Also a T cell Disorder
- Causes Recurrent Candidiasis

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PrepLadder

Immunodeficiency Disorders

Disorders of Phorgocytosis

1) Chronic Granulomatous Disease :
   - Deficiency of NADPH oxidase
   - Catalase positive organisms infections
2) LAD syndrome (Leukocyte adhesin deficiency)
   - Absence of CD 18
3) Chediak Higashi syndrome :
   - Defective phagolysosome fusion
   - Partial albinism
4) Job syndrome : TH – 17 deficiency
   ↑ IgE
5) MPO deficiency

T Cell disorders

1) Di George Syndrome :
   - Absence of thymus & parathyroid glands
2) Chronic mucocutaneous candidiasis

B – Cell Disorders

1) Bruton's X linked agammaglobulinaemia
   - Deficiency of Tyrosine kinase blocks B cell maturation
2) X-linked hyper IgM syndrome
3) Selective IgA deficiency
4) Common variable Hypogammaglobulinemia
5) Transient Hypogammaglobulinemia

Combined B & T cell disorders

1) Severe Combined immunodeficiency (SCID):
   - Cytokine receptor mutation
   - Adenosine deaminase deficiency
2) Wiskott Aldrich syndrome :
   - Recurrent infections, Eczema, thrombocytopenia
3) Ataxia telangiectasia

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PrepLadder

Unit: 7 General Microbiology

Chapter 18

History, Microscopy, Stains

Structure & Physiology of Bacteria

SCIENTIST IN MICROBIOLOGY

00:00:15

LOUIS PASTEUR

- Father of microbiology
- Proposed Fermentation principle
- Dis approved abiogenesis theory
- Proposed Germ theory
- Proposed Autoclave & Hot air oven
- Proposed pasteurization of milk
- Vaccines

  C → Cholera

  A → Anthrax

  R → Rabies → 1st rabies vaccine given to JOSEPH MEISTER

LOUIS PASTEUR

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ROBERT KOCH

ROBERT KOCH

- Father of modern Microbiology
- Proposed KOCH POSTULATES

PrepLadder

- Discovered TB, cholera bacilli
- Proposed Hanging Drop method
- Proposed Aniline dye staining
- Proposed solid media concept

- KOCH POSTULATES
  - MTB [example]
  - Constant association with disease must be present with causative organism
  - Isolation in a pure culture media should be possible
  - Culture growth inoculated in animal should cause same lesion
  - Reisolation should be possible From experimental animal
  - Antibodies should be produced from antigens

BACTERIA Does not Follow KOCH POSTULATES

- Mycobacterium leprae
- Treponema pallidum

PAUL EHRLICH

- Proposed Acid Fast staining
- Standardization of toxin & anti toxin done by him

JOSEPH LISTER

- Father of antiseptic surgery

ANTONY VON LEEUWENHOEK

- Father of microscopy
- Coined the term "Little Animalcules" For micro organism
- Father of vaccination → EDWARD JENNER
- Father of PCR → KARY B MULLIS
- KLEB LOFFLER'S BACILLI → Coryne bacterium diphtheria
- PREISS NOCARD BACILLI → Coryne bacterium pseudo TB
- KOCH WEEK BACILLI → Haenophilus aegypticus
- WHITMORE BACILLI → Burkholderia pseudomallei
- PFFIFFER' S BACILLI → Haemophilus influenza

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Charcterstics	Viruses	Bacteria	Fungi	Protozoa/ Helminths
Cells	No	Yes	Yes	Yes
Diameter [μm]	0.2-2	1-5	3-10 (yeast)	15-25 [trophozoite]
Nucleic Acid	DNA or RNA	DNA & RNA	DNA & RNA	DNA & RNA
Type of Nucleus	None	Prokaryotic	Eukaryotic	Eukaryotic
Ribosomes	Absent	70s [50s + 30s]	80 s [60s + 40s]	80s [60s + 40s]
Mitochondria	Absent	Absent	Present	Present
Outer surface	Capsid	Rigid wall Peptidoglycan	Rigid wall Chitin	Flexible Membrane
Motility	None	Some	None	Most
Replication	-	Binary Fission	Budding mitosis	mitosis

MICROSCOPY

00:22:43

- Depends on
  - Magnification
  - Resolution → ability to distinguish two points as separate

Human eye → 0.2 mm

Light microscope → 0.2 μ

Electron microscope → 0.2 nm

Contrast can be ↑ ed by staining

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PrepLadder

LIGHT MICROSCOPE

- White light is used as source
- used For
  - Motility [hanging drop method] denomonstration
  - Gram staining
  - Acid Fast staining
- Internal details cannot be demonstrated
- Highest magnification achieved by oil immersion lens [100x X 10x = 1000x]

PHASE CONTRAST MICROSCOPY

- light emerges in different phases

  ↓

  Difference in intensity

  ↓

  Reveal internal structures of living cells
- can be used for motility demonstration

DARK FIELD MICROSCOPY

- Background is dark by dark Field condensers
- Reflected light is used
- Visualise spirochaetes

FLUORESCENT MICROSCOPY

- When UV rays fall on fluorescent substance, wave length of invisible light increases & becomes Luminous
- STAINS

  Auramine / Rhodamine for → MTB

  Calcoflour White for → Fungus

  Acridine orange for → Malarial parasite

ELECTRON MICROSCOPE

- Beam of electrons used instead of Light
- Beam focused by circular magnets instead of lens

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PrepLadder

- Object is held in path of beam
- Scatters the electrons by object
- Image is formed on fluorescent viewing screen
- Maximum resolution can obtained [0.2nm-0.5nm] (100 times better than light microscope)
- Vacuum is used

Prokaryotes vs Eukaryotes

00:40:21

	PROKARYOTES	EUKARYOTES
Nuclear membrane	Absent	Present
Ribo nucleo protein	Absent	Present
Ribo nucleo protein	Absent	Present
Cell division	Binary Fission	Mitosis/meiosis
Chromosom	One, circular	Many, linear
Cell wall	NAM, NAG	Sterols [mycoplasma also]
Membrane bound organelles	Absent	Present
Respiration	Mesosomes	Mitochondria
Cytoskeleton	Absent	Present
Ribosomes	70s	80s
mRNA	No introns	Introns & Exons + nt

SHAPES

00:45:59

cocci in clusters → staphylococcus

cocci in chains → strepto coccus

cocci in pair [diplococcic] → Pneumococcus

Kidney shaped → Gonococci

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PrepLadder

Lens shaped	→	Meningococci
Cocci in Tetrads	→	Micrococci
Cocci in octate	→	sarcina
Spectacle like appearance	→	Enterococcus

BACILLI

Bamboo stick appearance	→	Bacillus anthracis
Diplo bacilli	→	Klebsiella pneumonia
Branching	→	Actinomycetes
Curved GNB	→	campylobacter
	→	Gullwing shape Helico bacter
Spiral	→	spirillum
		Spirochaetes
Pleomorphic	→	Haemophilus
		Brucella

All cocci are Gram positive Except

- Meningococci
- Gonococci
- Veilonella
- Moraxella

All Bacilli are Gram negative Except

M → Mycobacterium species
A → Anthracis Bacillus
C → Clostridium corynebacterium
D → Diphtheriae corynebacterium
O
N → Nocardia
A → Actinomycetes
L → Listeria
D → Diphtheroids

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Gram Positive vs Gram Negative Bacteria

00:55:18

	GRAM POSITIVE	GRAM NEGATIVE
THICKNESS	100 layer thick	1-2 layer thickness
LIPID CONTENT	2-5 %	15-50 %
TEICHOIC ACID	Present	Absent
VARIETY OF AA	Few	Several
LPS [Endotoxin]	Absent	Present
NAG-NAM		
Attached with a cross linked	Tetrapeptide side chain by pentapeptide crossbridge	Tetra peptide are directly linked to each other

NAG → N acetyl Glucosamine

N acetyl Muramic Acid

AA → Amino Acids

CELL WALL DEMONSTRACTION

R → Reaction with specific antibody

M → Micro dissection, Mechanical Rupture of cell

P → Plasmolysis

Doctors → Differential staining

INCLUSIONS

01:02:43

STORAGE GRANULES

- Corynebacterium diphtheria
  - Polymeta phosphate
  - Volutin granules
  - Meta chromatic granules
  - Babes ernest granules

- Energy deposits of bacteria
- Granules are staines by

P → PONDER'S STAIN

A → ALBERT STAIN

N → NEISSER STAIN

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CAPSULE

01:05:00

- Polysaccharide mostly
  Polypeptide in Bacillus anthracis
- Virulence factor
  prevents phagocytosis
- Demonstrated by
  Negative staining for crytococcus
  Quellung Reaction for Pneumococcus
  Mc Fadyean's Reaction

FLAGELLA

01:06:54

- Used for motility
- 0.02 μ thick
  3-20 μ long
- Antigenic
- PARTS
  Filament
  HOOK
  Basal Body → attached to cytoplasmic membrane by rings
  - T → Tumbling → Listeria
    D → Darting → Vibrio
    S → Swarming → Proteus

  Falling leaf like → Giardia lamblia [d/t 4 pairs of Flagella]
- WIDAL TEST → measure antibodies against 'O' & 'H' [more immunogenic]

FIMBRIA

01:11:06

- Fine hair like structure
- 0.5 μ long
- helps in adhesion
  Conjugation [sex pili]
- Antigenic

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PrepLadder

SPORES

01:12:28

- Highly resistant resting Forms
- Spore forming Bacteria
  - B → Bacillus anthracis
  - S → Sporo sarcina
  - C → clostridium
    - → Drum stick appearance seen in cl. Tetani
    - → most have sub terminal spores [most of clostridia]
    - → central spore seen in cl. biFermentans

PROTOPLAST → seen in Gram Positive

SPHEROPLAST → seen in Gram negative

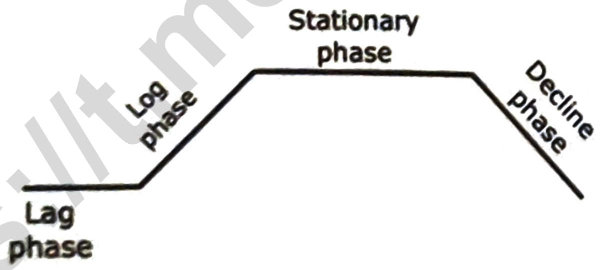
Without cell wall Forms [looses cell wall in Hypertonic solution]

L FORMS

- Cell wall free forms
- KLINBERGER named them
- Seen in Streptobacillus moniliformis
- Unstable L forms seen in Mycoplasma

GROWTH CURVE

01:17:40



- Lag Phase
  - Build up enzymes & metabolites
  - Adopts to the conditions → ↑ in Size
- Log Phase
  - Exponential division occurs
  - Maximum active
  - Uniformly stained
  - $(2)^n$ exponential division

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- Stationary Phase
  - Live = Dead
  - Sporulation happens
    Exotoxin production happens
    Antibiotic production [some bacteria] happens

- Decline Phase
  - Complete accumulation of toxic metabolites
  - Involution forms Formed

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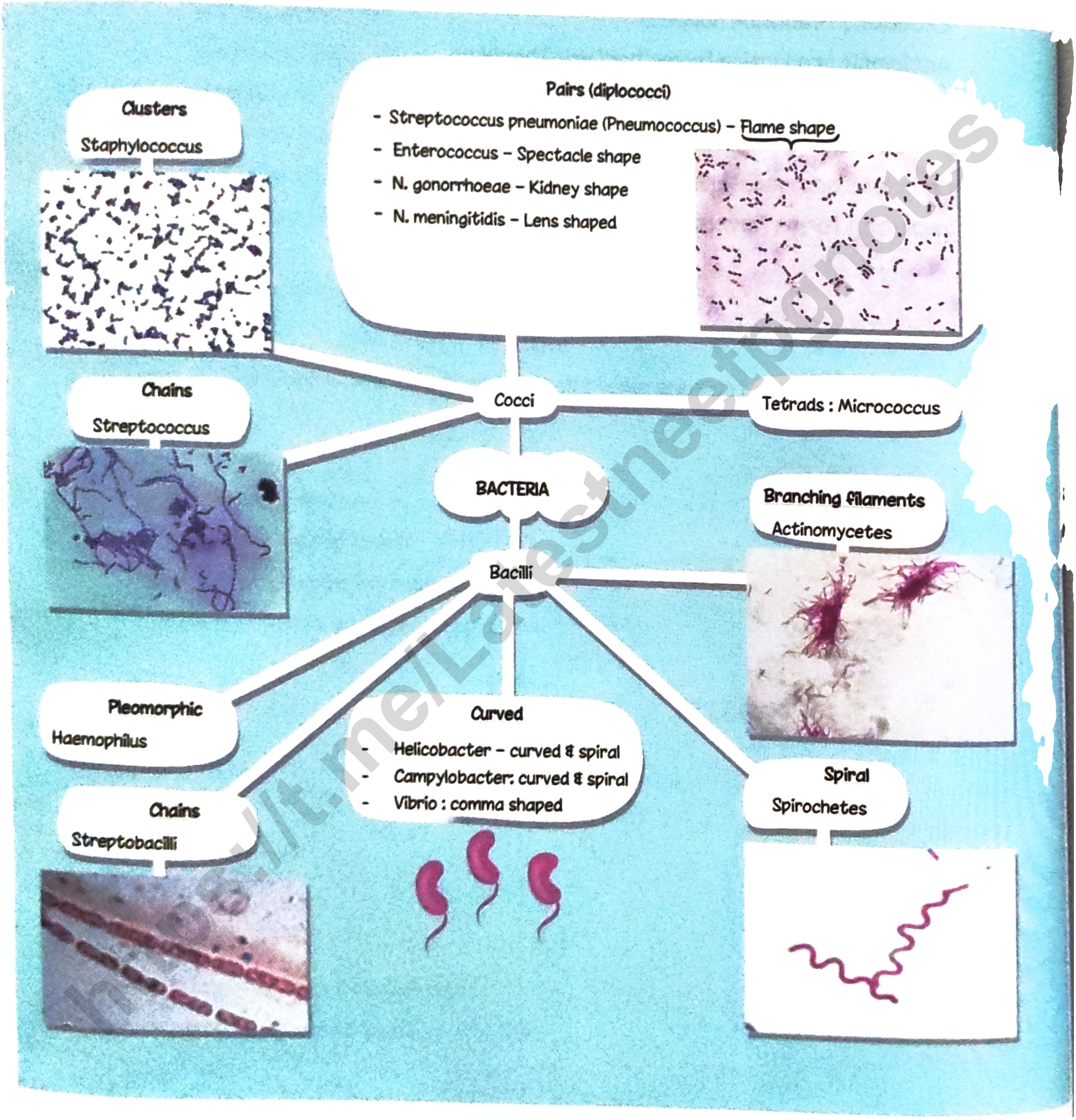
PrepLadder



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Chapter 19

STERILIZATION & DISINFECTION

STERILIZATION

00:00:30

- Making an article free of pathogens, non pathogens including spores

DISINFECTION

- Removal of pathogens & non pathogens but not spores

ASEPSIS

- Process in which the chemical agents [antiseptic] applies to body surfaces will Kill/inhibit pathogenic microorganisms

DECONTAMINATION OR SANITIZATION

- Reduction of Pathogenic microbial population to a level in which items are considered safe to handle

NATURE OF ORGANISMS [RESISTANCE IN DECREASING ORDER]

- Prions › cryptosporidium oocysts › Bacterial spores › mycobacteria › other parasite cysts [eg. Giardia] › small non-enveloped virus › Trophozoites › Gram negative bacteria › Fungi › large non-enveloped viruses› Gram positive bacteria › Enveloped viruses

CLASSIFICATION OF STERILIZATION METHODS

1. SUN LIGHT
2. DRYING
3. HEAT

a. DRY HEAT
- Flaming
- Incineration
- Hot air oven

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b. MOIST HEAT

< 100°c	Pasteurization Inspissation Water bath
= 100°C	Tyndallisation Boiling
> 100°C	Autoclave

4. FILTRATION — Depth Filters, Membrane filters

5. RADIATION
   - a. IONISING — Gamma rays & x Rays
   - b. Non ionizing — UV Rays & infrared rays

6. ULTRASONIC VIBRATION

7. CHEMICALS — Alcohols, Aldehydes, Phenolic compounds Biguanides, Halogens, Oxidising agent, Heavy metal salts, Surface active agents, dyes

8. GAS STERILIZATION — ETO, BPL, Plasma sterilization

METHOD	PRINCIPLE
1 SUN LIGHT	Active microbicidal activity of Light UV LIGHT
2 DRYING	70-80% weight of bacterial cell is water
3 DRY HEAT	C → Charring O → Oxidative damage D → Denaturation E → Elevated electrolytes
4 MOIST HEAT	Denaturation & Coagulation of Proteins
5 FLAMING	Flame of BUNSEN BURNER Inoculating Wires, 100ps
6 INCINERATION	870°C to 1200°C burns the anatomical & microbiological waste

HOT AIR OVEN

00:03:35

- Temperature → 160°C for 2 hrs
- Electrically heated chamber with a Fan to ensure adequate & even distribution of hot air

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- **Material sterilized**
  - Glassware → Syringes, petridishes, Flasks, pipettes
  - Surgical instruments → Scalpels & scissors
  - Chemical like → Dusting powder, grease, paraffin, oils

- **PRECAUTIONS TO BE TAKEN**
  - Overloading should not be done
  - Equal & Free air circulation should be maintained
  - Material should be properly dried before putting
  - Paper wrapping of items should be done
  - Oven should be allowed to cool For 2 hrs before opening the door

- **CONTROLS**

  1) Spores of Bacillus subtilis

  2) Spores of clostridium tetani [non pathogenic]

MOIST HEAT STERILIZATION

00:07:21

- Coagulation & denaturation of proteins
- < 100°C → Pasteurization, inspissations

  at 100°C → Boiling, Tyndallisation

  > 100°C → Autoclave

PASTEURIZATION OF MILK

00:09:07

Flash method → 72°C For 15-20 sec → bring down to 13°C
→ Kills coxiella burnetti spores also
→ Better method than Holder's

Holder's method → 60°C for 30 min

WATERBATH

- Disinfection of serum, Body Fluids & vaccines can be sterilized
- Bacterial vaccines → disinfected at 60°C x 1 hr

  Serum or heat labile body Fluids → disinfected at 56°C x 1hr

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INSPISSATION

00:10:55

- 80-85°C for 20-30 minutes for 3 days
- First exposure → vegetative forms Killed

  2nd 3rd exposure → spores germinating also get Killed
- Done For
  - LJ [LOWENSTEIN JENSEN] MEDIA / DORSET EGG MEDIA
  - LSS [LOEFFLER'S SERUM SLOPE For Diphtheriae]

AT 100°C

BOILING

- 100°C for 15 minutes
- most of vegetative Forms Killed
- but SPORES are NOT KILLED

KOCH'S ARNOLD STEAM STERILIZER

- Used For those medias which decompose at higher temperature of autoclave
- Articles exposed to steam [100°C] at atmospheric pressure For 90 min

TYNDALLIZATION

00:12:42

- 100°C x 20-30 min x 3 Days
- Done For Sugar & Gelatin containing Substances

AT > 100°C

AUTOCLAVE

00:13:25

- WATER BOILS WHEN ITS VAPOUR PRESSURE EQUALS SURROUNDING ATMOSPHERIC PRESSURE
- As atmospheric pressure increases, Boiling temperature also increases
- At normal pressure, boiling point of water is 100°C

3 PARTS OF AUTOCLAVE

a) PRESSURE CHAMBER

b) Lid

c) Electrical heater

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TEMPERATURE OF AUTOCLAVE

- 121°C x 15 min x 15 psi [Pounds square inch]
- 126°C x 10 min x 20 psi
- 133°C x 3 min x 30 psi

DONE FOR

- Surgical instruments
- All culture medias except LJ & LSS | Dorset egg
- Autoclavable plastic containers
- Plastic tubes & pipette tips
- Biohazard waste
- Surgical Sutures except catgut

CONTROLS → Spores of GeoBacillus stearothermophilus
→ Killed at 121°C x 12 min

CHEMICAL INDICATORS

- Class I → External pack control [Autoclave tape]
- Class II → Equipment control [Bowie dick Test]
- Class IV/V → Internal pack control

PHYSICAL CONTROL

- Digital displays on equipment temperature, pressure & time

FILTRATION

00:16:42

- Mainly done For Heat sensitive substances
  - Vaccines
  - Antibiotics
  - Serum
  - Toxin
  - Sugar solution

2 TYPES OF FILTERS

1) **DEPTH FILTERS** – POROUS FILTERS [metal polymer with random mats]

- Eg
  - Candle Filters
  - Unglazed porcelain Filters

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- Seitz Filter
- Sintered glass filter

2) MEMBRANE FILTERS

- Most widely used filters for bacterial Filtration
- Made up of cellulose acetate, cellulose nitrate, polycarbonate
- Pore size → 0.22 μ → mc used
  → 0.45μ → retains coliforms
  → 0.8 μ → retains airbone micro organisms
- USE → to sterilize sugar, sera & antibiotic solutions

AIR FILTERS

(1) HEPA [High Efficiency Particulate Air] FILTER

- Removes 99.97% of particles having a size 0.3 μ or more

(2) ULPA [Ultra Low Particulate/Penetration]

- Removes 99.999% of dust, pollen, molds

CONTROL → Brevundimonas diminuta & serratia marcesens

RADIATION 00:19:28

IONIZING RADIATION / COLD STERILIZATION

- Gamma rays / x Rays
- Breaks upon DNA without temperature rise → COLD
- Used to sterilize plastic syringes & catgut sutures
- Control → Spores of Bacillus pumilus

NON IONIZING RADIATION / HOT STERILIZATION

- UV rays / Infra red rays
- Temperature of surface increases
- used For OTs, Biosafety cabinets

HIGH FREQUENCY ULTRASONIC WAVES

- Disrupt the bacterial cells
- Not reliable, not done these days

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CHEMICAL METHODS OF STERILIZATION

00:22:23

VARIOUS GROUPS OF DISINFECTANTS

ALCOHOLS

- Most widely used disinfectants & antiseptics

ETHANOL & ISOPROPONAL [70-80%]

- MOA → Bactericidal & Fungicidal but not sporicidal
  → Some enveloped virus like HIV are also destroyed

ETHYL ALCOHOL → used as surgical spirit [70%]
→ used for antirubs as antiseptics

ISOPROPYL ALCOHOL → used for stethoscope & thermometer x 10-15 min

ALDEHYDES

- MOA
  - Combine with nucleic acids, proteins & inactivate them by cross linking & alkylating the molecules
  - Also sporicidal [used as chemical Sterilants]

FORMALDEHYDE

- FORMS

1) GAS → used For OT Fumigation
2) LIQUID [FORMALIN] → used to preserve anatomical & patho specimens

GLUTARALDEHYDE

- Less toxic, less irritant
- 2% Gluteraldehyde → CIDEX
  - Used For Bronchoscopes, cysto SCOPES, endoscopes
- Disinfects objects within 20 min
- Available in inactive form & activated by alkalinisation before use
- Remains active only For 14 days

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ORTHOPHTHALDEHYDE

- 0.55% Solution can also be used For sterilizing cytoscopes & Endoscopes

Advantages

- Does not require activation
- Low vapour property
- Better odour
- More stable
- ↑ mycobactericidal activity

PHENOLIC COMPUNDS

PHENOL [CARBOLIC ACID]

- 1st Widely used antiseptic
- Discovered by JOSEPH LISTER in 1867
- Derived from coal tar
- Mechanism → denature proteins & disrupts cell membranes
- Advantage → high mycobactericidal activity & can act upon in pressure of organic waste
- CRESOLS, XYLENOL, LYSOL are used as disinfectants in labs & Hospitals
- Retain activity in the presence of organic waste

CHLORHEXIDINE → components of SAVLON [chorhexidine + cetrimide]

CHLOROXYLENOL → component of dettol

HALOGENS

- Iodine & chlorine have antimicrobial activity
- Exist in free state & Form salt like compounds with Sodium & most other metals

IODINE

- Skin antiseptic
- oxidizing cell constituents & iodinizing cell proteins
- at higher concentration, it may kill spores

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TINCTURE OF IODINE [2% IODINE]

- IN WATER Ethanol Solution of Potassium Iodide

IODOPHOR

- Complexing iodine with organic carrier → BETADINE

CHLORINE

COMMON ACTIONS

1) Bleaching in laundry & homes by oxidation
2) Disinfection in hospitals by coagulation
3) Odour control in toilets by precipitation
4) Chlorination of drinking water in water tanks by septication
5) Elimination of slime & algae in Swimming pool by PH adjustment

PREPARATIONS

1) Chlorine Gas 2) Sodium Hypochlorite 3) Calcium Hypochlorite	Produce Hclo [Hypochlorous Acid] ↓ Oxidation of cellular materials & destruction of vegetative bacteria & fungi

- Organic matter interferes with its action

OXIDIZING AGENTS

H_2O_2
Peracetic Acid
Plasma Sterlization

H_2O_2

- Strong oxidizer [high level disinfectants] Liberates toxic free hydroxyl radicals which are active ingredients
  Attack membrane, DNA, lipid & other cellular components
- Most effective concentration → 3-6%
  Spores might require → 10% [higher concentration]

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PERACETIC ACID

- Powerful oxidizing agent
- More active than H2O2
- Concentration → < 1% is sporicidal

At low temperature

- High level disinfectant
- Used to sterilize endoscopes
- May corrode steel, iron, copper

PLASMA STERILIZATION

00:31:59

- Plasma Refers to gaseous State consisting of ions & free electrons
- Chemical Sterilants H_2O_2 & Peracetic acid used to induce Plasma state
- Plasma sterilizers [STERRAD] → maintains uniform vaccum inside the chamber
- Active agents → UV photons & radicals used
- maintain low temperature
- used in sterilization of surgical instruments
- CONTROL → Bacillus Stearothermophilus spores

HEAVY METAL SALTS

MECHANISM OF ACTION

- Combine with bacterial cell proteins, often with their sulfhydryl groups & inactivate them & precipitate cell proteins
- Most heavy metals are bacteriostatic
  - SILVER SULFADIAZINE [used For Burns]
  - SILVER NITRATE [1%]
  - COPPER SULPHATE [Fungicidal]
  - MERCURY SALTS
    - MERCUROCHROME → Known antiseptics
    - THIOMERSAL → preservatives in vaccine & sera

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SURFACE ACTIVE AGENTS OR SURFACTANTS 00:36:10

- Lower surface tension b/w 2 liquids or b/w liquid & solid
- CLASSIFIED INTO
  - Anionic
  - Cationic
  - Non ionic
  - Amphoteric

CATIONIC SURFACTANTS

- Quaternary Ammonium compounds are most popular cationic disinfectants
- Have positively charged quaternary nitrogen & a long hydrophobic aliphatic chain
- They disrupt the microbial membranes & denature the proteins
- Gram positive bacteria are better Killed than gram negative bacteria but not MTB or spores
- Cationic detergents → disinfectant For Food utensils & small instruments
- Eg→ Alkyl trimethyl ammonium salts
  Acetyl trimethyl ammonium bromide [cetrimide]

ANIONIC SURFACTANTS [common soaps]

- Have strong detergent action
- Have weak antimicrobial action
- Most active at acidic PH

AMPHOTERIC [Tego compounds]

- Have detergent property of anionic compounds
- Have Antimicrobial property of cationic compounds
- Act upon compounds at a wide range of PH but activity reduced in presence of organic waste
- Eg. TEGO compounds → Active compounds is DDAG [Dodecyl Diamino Ethyl Glycine]
- Used as a antiseptic in dental practices

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DYES

- ANILINE & ACRIDINE are 2 groups of dyes
- Used as skin & wound antiseptics

ANILINE DYES

1) CRYSTAL VIOLET
2) GENTIAN VIOLET
3) MALACHITE GREEN
   - More active against Gram positive than Gram negative
   - But no activity against MTB
   - Reduced activity in presence of organic waste
   - Interferes with synthesis of peptidoglycan in cell walls

ACRIDINE DYES

ACRIFLAVINE
EUFLAVINE
PROFLAVINE
AMINACRINE

} affected very little by organic waste

- MORE ACTIVE AGAINST Gram positive than Gram negative
- Interfers in synthesis of Nucleic Acid & proteins in bacterial cells

GASEOUS STERILIZATION

- Ethylene oxide [ETO]
- Low temperature steam formaldehyde
- propiolactone

ETO

- Most widely used gaseous chemical sterilant
- Both microbicidals & sporicidal
- Acts by combining with cell proteins
- High penetrating power
- Used For pre packed syringes
- Highly inflammable, irritant & explosive

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- Mixed with CO_2 or dichlorodifluoromethane

3 Factors affecting ETO disinfectant power

a) ETO concentration

b) Humidity

c) Temperature

At ETO concentration of 700 mg/l & 40-50% relative humidity at 38°C, sterilization is achieved [5-8hrs]

CONTROL → Bacillus globigi spores used

USES

- to sterilize
  - Disposable plastic petri dishes
  - Syringes
  - Heart Lung machines
  - Respirators
  - Sutures
  - Catheters
  - Dental equipments

LOW TEMPERATURE STEAM FORMALDEHYDE

- Used For Fumigation of Ots
- Not used now

BPL [Beta PropioLactone]

- 0.2% is active against all micro-organisms including spores
- Used For inactivation of vaccines
- Found to be carcinogenic, so not used for Fumigation

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TESTING OF DISINFECTANTS

00:40:10

PHENOL COEFFICIENT TEST/RADIAL NAEKER TEST

- Determined by dilution of disinfectant
- Used to sterilize suspension of salmonella typhi in given time divided by dilution of phenol which sterilizes suspension in same time
- If Phenol coefficient › 1 → disinfectant is better than phenol

- PHENOL COEFFICIENT TEST =

$$\frac{\text{Highest dilution of test disinfections that kills S. typhi in given time}}{\text{Highest dilution of phenol that Kills S. typhi in given time}}$$

- It doesn't assess activity in presence of organic waste

CHICK MARTIN TEST

- Modification of RIDEAL WALKER CO EFFICIENT TEST
- Checks Whether Disinfectant works in presence of organic waste [faeces, yeast] to simulate natural condition

CAPACITY TEST [KELSEY SYKES TEST]

- Capacity of a disinfectant to retain its activity when repeatedly used microbiologically in increasing load

IN USE TEST [KELSEY & MAURER TEST]

- Disinfectant is microbiologically contaminated
- Routinely performed in hospitals once in 3 months

SPORICIDAL AGENTS

E → Ethylene oxide
F → Formaldehyde
G → Glutaraldehyde [2%]
H → Hydrogen peroxide
→ Sodium Hypochlorite [blood spills]
O - Ophthalic Acid
Peracitic acid
Autoclave
Hot air oven
Plasma sterilization

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CHEMICAL DISINFECTANTS – CLASSIFICATION

Levels of disinfectants	Bacterial spores	TB bacilli	Non enveloped virus	Fungi	Enveloped virus	Vegetative bacteria
Low level	No	No	No	+/-	Yes	Yes
Intermediate level	No	Yes	Yes	Yes	Yes	Yes
High level	May be	Yes	Yes	Yes	Yes	Yes
Chemical sterilant	Yes	Yes	Yes	Yes	Yes	Yes

SPAULDING CLASSIFICATION

CRITICAL DEVICES

- Includes those Enters sterile sites like Surgical instruments, catheters
- High level disinfection needed

SEMICRITICAL DEVICES

- Includes those comes in contact with mucous membranes like respiratory equipments [laryngoscope]
- High level disinfections needed

NON- CRITICAL DEVICES

- Includes those come in contact with skin like BP cuff, ECG Electrodes
- Low/intermediate level disinfection needed

MEDICAL EQUIPMENTS

- Includes Examination tables, computers
- Low level disinfections needed

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Chapter 20

BACTERIAL GENETICS

DNA

- made of
  - Deoxy ribose
  - Phosphate groups
  - 4 nitrogenous bases
    - 2 purines → Adenine & Guanine
    - 2 pyrimidines → Thymin & cytosine
  - Strands held together by hydrogen bonds

RNA

- made up of
  - Ribose sugar
  - Uracil replaces the Thymine

- Types
  - mRNA [messenger RNA]
  - tRNA [Transfer RNA]
  - rRNA [Ribosomal RNA]

GENE → Segment of DNA that specifies for a particular polypeptide

CODON → Genetic information stored in DNA

ANTICODON → Binds to codon

NON SENSE CODON → UAA, UAG, UGA
→ DO not code for any aminoacid
→ Terminate the synthesis of polypeptide
→ aKa STOP CODONS

EXTRA CHROMOSOMAL GENETIC MATERIALS 00:06:02

PLASMIDS

- Free in cytoplasm of bacteria
- When they integrate with host cell → EPISOME

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- Not essential For the Survival of bacteria
- Plasmid Encode genetic information for
  - Resistance to antibiotics
  - Bacteriocins
  - Enterotoxins
  - Enhanced pathogenicity
  - Degrading complex organic molecules

BACTERIAL VARIATIONS

00:09:43

PHENOTYPIC	GENOTYPIC
Physical expression in environment	Sum total of gene make up ↓ Genetic Apparatus of the cell
Examples (1) TYPHOID BACILLI – NORMALLY flagellated – No Flagella grow in phenol agar (2) LACTOSE FERMENTATION by E.coli – By β galactosidase	Examples (1) Mutations (2) Transfer of genes by – Conjugation – Transformation – Transductions

MUTATION

00:14:14

- Random, undirected, heritable variation
- Alteration of nucleotide sequences happens
  By Addition
  Deletion
  Substitution [one or more bases]
- Bacteria undergo mutations → 10^{-2} to 10^{-10} per bacterium per division
- MUTAGENIC AGENTS
  - UV Rays
  - Alkylating agents
  - Acridine dyes

- All genes are susceptible For mutations
  But all mutations are not expressed

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- LETHAL MUTATION → Harmful, Destroys the vital function
- CONDITIONAL LETHAL MUTANTS

  Example → Temperature sensitive mutants

  - Can lives at 35°C
  - But dies at 39°C [Restrictive temperature]

- TYPES

1) Point mutation

   Base pair substitution

   Frame shift mutation

2) Multisite Mutations

   Gain, Loss or Duplication can occur

POINT MUTATION

00:21:30

TRANSITION	TRANSVERSION
Mc type Purine is replaced by purine Pyramidine is replaced by pyramidene	Purine can be replaced by Pyramidine & VICE VERSA

FRAME SHIFT MUTATION

			(serine)	
Normal	→	AAT	ACT	GCC
Deletion	→	AAT	CTG	CCA
			(valine)	

Normal	→	AAT	ACT	GCC
Insertion	→	AAT	CACT	GCC
			(Glutamine)	

MULTI SITE MUTATIONS

00:25:53

TYPES

1) Addition/ Gain
2) Deletion/ Loss
3) Duplication
4) Inversion

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MISSENSE MUTATIONS

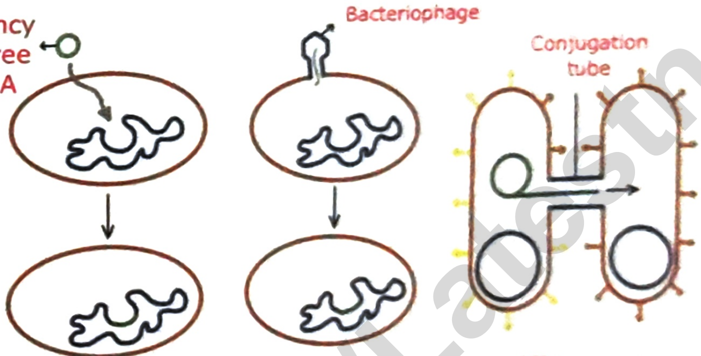
- Triplet code is altered, different amino acid from that normally Located at that particular position

NONSENSE MUTATIONS

- Premature polypeptide chain termination by stop codon

GENE TRANSFERS [sexual process] 00:28:58

1) TRANSFORMATION → naked DNA taken up from the environment by bacterial cells
2) TRANSDUCTION → use of bacteriophages [bacterial virus] to transfer DNA b/w cells
3) CONJUGATION → direct transfer of DNA from one bacterial cell to another



TRANSFORMATION 00:31:48

- Discovered by GRIFFTH
  by AVERY, MACLEOD & Mc CARTY [1994]

- CASE A
  Living non-encapsulated bacteria [Rough strains of Pneumococcus when injected into mouse → mouse dies

- CASE B
  Heat Kills encapsulated pneumococcus
  When injected into mouse → No effect

- CASE C
  Heat Killed smooth strain of Pneumonia + Live rough strains of Pneumonia

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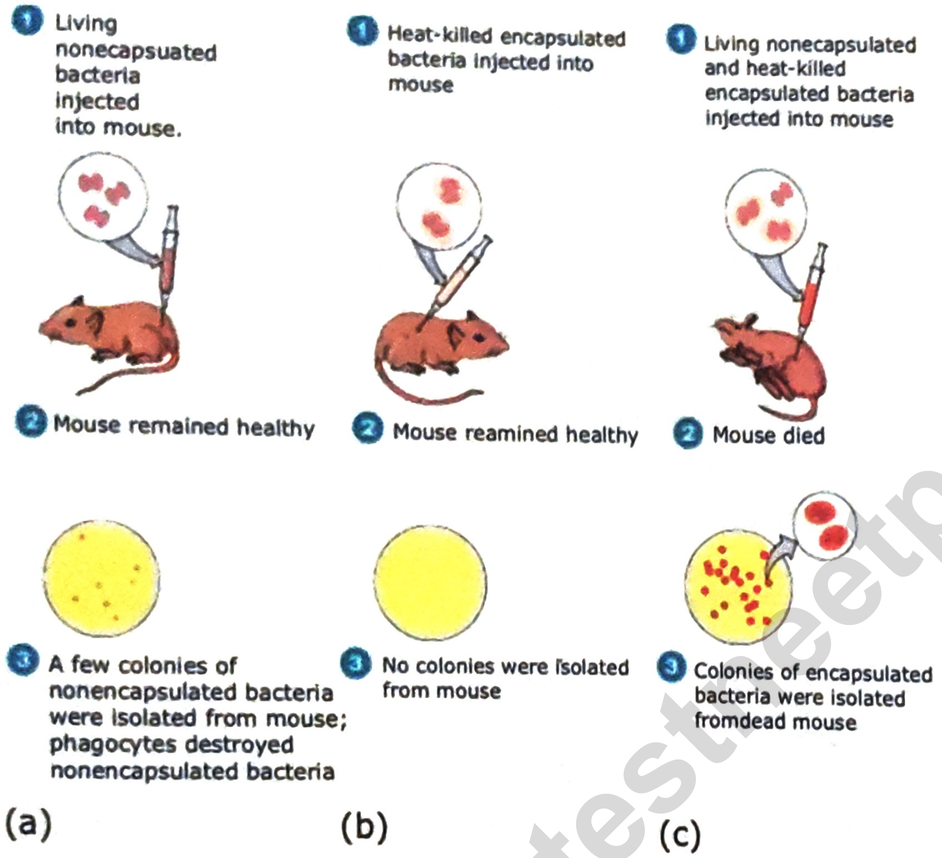
When injected into mouse → Mouse dies

Colonies of living encapsulated pneumococci are isolated from dead mouse [through agency of free DNA]



Demonstration of Transformation by Avery, Macleod & Mc carty

TRANSDUCTION 00:34:49

- Transmission of portion of DNA From one bacterium to another by a bacteriophage
- MECHANISM
  - Part of host DNA may accidentally incorporate into bacteriophage
  - Gets transferes to recipient bacterium so that it leads to acquisition of new characters by the recipient bacterium from donor DNA

TYPES 00:42:33

LYTIC OR VIRULENT CYCLE

- Bacteriophage multiplies in host cytoplasm & produce large no. of daughter phages
- Subsequently gets released, causing death & lysis of host cycle

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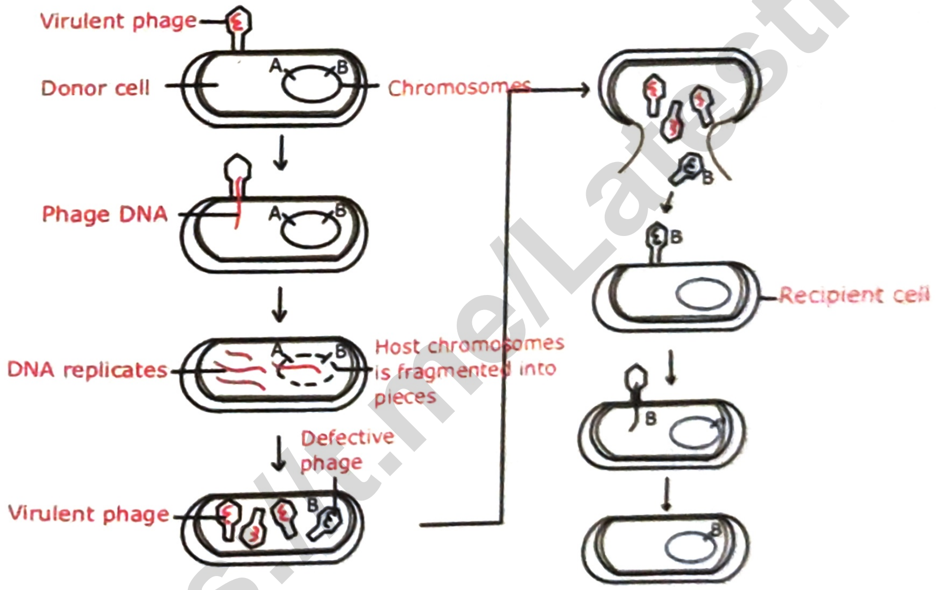
LYSOGENIC OR TEMPERATE CYCLE

- Host bacterium unharmed
- Phage DNA integrate with bacterial chromosome as prophage that multiplies Synchronously with bacterial DNA
- When phage DNA tries to come out it is disintegrated From host chromosome, comes out in cytoplasm & behaves like a lytic phage;
- It replicates to produce daughter phages which subsequently released by host cell lysis

TYPES OF TRANSDUCTION

GENERALISED TRANSDUCTION

- Involves transfer of any part of donor bacterial genome to recipient bacteria
- Occurs as a result of defective assembly during the lytic/virulent cycle & sometimes temperature phages



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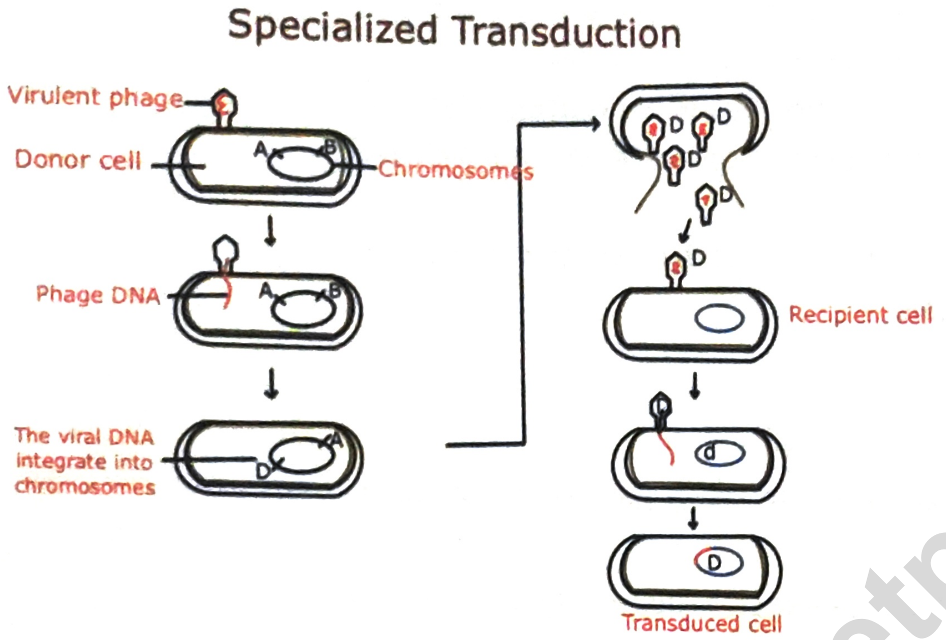
SPECIALIZED / RESTRICTED TRANSDUCTION

- Part adjacent to integration of phage DNA with host cell DNA



LYSOGENIC CONVERSION

- Occurs mostly in temperature / Lysogenic life cycle
- Phage DNA incorporates in Bacterial DNA & gives it a new property → LYSOGENIC CONVERSION

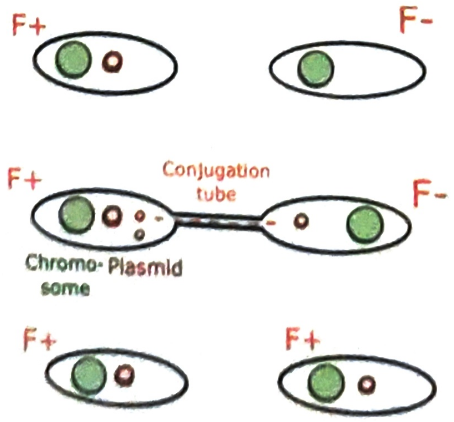
BACTERIAL TOXINS THAT ARE CODED BY LYSOGENIC PHAGES

A → A & C Pyrogenic toxins of Strepto Pyogenes
B → Botulinum toxin C & D
C → Cholera toxin
D → Diphtheria toxin
E → EHEC [Entero Haemorrhagic E.coli] [aka vero Toxin]
Panton valentine toxin of staph avreus

CONJUGATION

00:55:49

- MC method of resistance transfer in lysogenic bacteria
- Possibility 1



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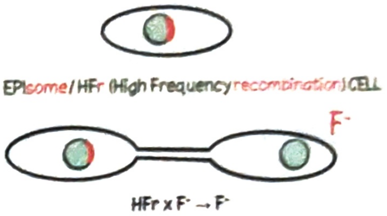
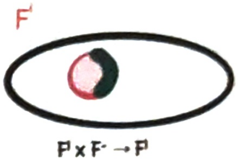
POSSIBILITY 2 / EPISOME / HFR [High Frequency Recombination] CELL



POSSIBLITY 3



POSSIBILITIES

F^+	x	F	→	F^+
HFr	x	F^-	→	F^-
F'	x	F^-	→	F'

TRANSFERABLE DRUG RESISTACE

01:00:32

- Example of conjugational transfer
- RTF + r = RESISTANCE
- RTF – Resistance transfer Factor
- r – Resistant determinant [main Factor]
- Leads to MDR [multidrug resistance] → difficult to treat

- MUTATIONAL DRUG RESISTANCE → SINGLE DRUG RESISTANCE
  → EASY TO Rx
- TRANSFERABLE DRUG RESISTANCE → MULTI DRUG RESISTANCE → DIFFICULT TO Rx

- In MTB, mc method of resistance is Mutational drug resistance

TRANSPOSONS/JUMPING GENES

01:02:37

- They are responsible for intracellular transfer
- Transfer happens b/w
  - Chromosome - chromosome
  - Chromosome - Plasmid
  - Plasmid - Plasmid

- Work in a cut & paste manner
- Have inverted repeat sequences & insertion sequence
- Method is Known as TRANSPOSITION
- Resistance is transferred mainly with transposons

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Chapter 21

BIOTERRORISM AGENTS

CATEGORY A → Carry highest risk
CATEGORY B
CATEGORY C → Emerging Threats

CATEGORY A

00:01:10

- Play → Plague
- Vith → Viral Haemorrhagic Fevers [Marburg, Ebola, KFD, Yellow Fever]
- Small → Small Pox
- B → Botuism
- A → Anthrax
- T → Tularemia

CATEGORY B

00:03:37

- C → Clostridium perferingers [Epsilon toxin]
- B → Brucella
- S → Salmonella, Shigella, Staphylococcus [enterotoxin B]
- E → E.coli [O_{157} : H_7]

- Vill → Vibrio cholera
- Take → Typhus Fever
- Questions For → Q Fever
- Medical → Meliodiosis
- P → Psittacosis
- G → Glanders [non motile]

CATEGORY C

00:10:06

- N → Nipah
- H → Hanta Virus
- S → SARS Corona Virus
  SARS COV-2

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Unit: 8 Clinical Microbiology

Chapter 22

NORMAL MICROBIAL FLORA

00:00:13

Introduction

- Normal flora is the term used to describe the various bacteria and fungi that are permanent residents of certain body sites, especially the skin, oropharynx, colon, and vagina.

- Viruses and parasites, which are the other major groups of microorganisms, are usually not considered members of the normal flora.

- Other names: commensals, human microbiome, indigenous microbiota

- The members of the normal flora vary in both number and kind from one site to another

- The skin and mucous membranes always harbor a variety of microorganism that can be arranged into two groups:

1) Resident microbiota: It consists of relatively fixed area at a given age; if disturbed, it promptly reestablishes itself for example E.coli in intestine.
2) Transient microbiota: It consists of nonpathogenic or potentially pathogenic microorganisms that inhabit the skin or mucous membranes for hours, days, or weeks
   Example MRSA.

- Normal flora depends upon various factors:
  1) Temperature
  2) Moisture
  3) pH
  4) Environment (community or hospital)

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5) Immune status

6) Anatomical sites

7) O_2

ROLE OF NORMAL FLORA

00:04:06

- **Beneficial effects:**
  1) Prevent colonization of pathogenic bacteria by competing for attachment sites and nutrients
  2) Synthesize vitamins (B,K) which are absorbed as nutrient
  3) Inhibits the growth of pathogenic bacteria by secreting waste substances (fatty acids, peroxidase, lactic acid, bacteriocin etc.)
  4) Immune stimulation
  5) Prevent allergic diseases (Hygiene hypothesis)
  6) Complement activation by endotoxin of normal flora

- **Harmful effects**
  1) Agent of disease
  2) Transfer to susceptible host
  3) Bacterial syngergism
  4) Contribute in spread of drug resistance
  5) Competition for host's nutrients

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00:09:54

Location	Important Organisms[1]	Less Important Organisms[2]
Skin	S. epidermidis	S. aureus, Corynebacterium Various Strept. Anaerobes Yeasts
Nose	S. aureus[3]	S. epidermidis Corynebacterium Various strept.
Mouth	Viridans streptococci	Various strept. Eikenella corrodens
Dental plaque	Strept. mutans	Prevotella intermedia Porphyromonas gingivalis
Gingival crevices	Various anaerobes (e.g., Bacteroides, Fusobacterium, Strept., Actinomyces)	
Throat	Viridans streptococci	Various strept. Neisseria species H. influenzae S. epidermidis
Colon	Bacteroides fragilis, E. coli	Bifidobacterium Eubacterium Fusobacterium Lactobacillus Various aerobes Gram -ve Rods Enterococcus faecalis Various strept.
Vagina	Lactobacillus, E. coli[3], group B strept.[3]	Various strept. Various gram -ve rods Corynebacterium C. albicans
Urethra		S. epidermidis Corynebacterium Various strept. Gram -ve rods

[1]Organisms that are medically significant or present in large numbers.

[2]Organisms that are less medically significant or present in smaller numbers

[3]These organisms are not part of the normal flora

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PROBIOTICS

00:19:00

- Probiotics are live, nonpathogenic bacteria (or yeasts) that may be effective in the treatment or prevention of certain human diseases
- They are useful in the condition where the normal intestinal flora is suppressed.
- Examples: Bacillus coagulans, Bifidobacterium longum, Lactobacillus acidophilus, Saccharomyces boulardii.

PREBIOTICS

00:20:20

- In contrast to probiotics, prebiotics are the dietary non-digestible fibres which when administered, stimulates the growth and activity of normal flora.

Q1. The colon is the site of the largest number of normal flora bacteria. Which one of the following bacteria is found in the greatest number in the colon? 00:20:56

(A) Bacteroides fragilis

(B) Clostridium perfringens

(C) Enterococcus faecalis

(D) Escherichia coli

A. (A) Bacteroides fragilis

Q2. Certain microorganisms are never considered to be members of the normal flora. They are always considered to be pathogens. Which one of the following organisms fits into that category? 00:21:28

(A) Streptococcus pneumoniae

(B) Escherichia coli

(C) Mycobacterium tuberculosis

(D) Staphyloccus aureus

A. (C) Mycobacterium tuberculosis

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PrepLadder

Q3. Your patient is a 30-year-old woman with a previous history of rheumatic fever who has had fever for the past 2 weeks. On examination, you find a new heart murmur. Your suspect endocarditis and do a blood culture, which grows a viridians group streptococcus later identified as S. sanguis. Using your knowledge of normal flora, what is the most likely sources of this organism?

00:22:00

(A) Duodenum

(B) Skin

(C) Throat

(D) Urethra

A. (C) Throat

Q4. A 76-year-old woman with a prosthetic (artificial) hip comes to you complaining of fever and pain in that joint. You are concerned about an infection by S. epidermidis. Using your knowledge flora, what is the most likely source of this organism?

00:23:25

(A) Dental plaque

(B) Mouth

(C) Skin

(D) Stomach

A. (C) Skin

Q5. Antimicrobial therapy can decrease the amount of susceptible bowel flora and follow proliferation of relatively resistant colonic bacteria. Which one of the following species can proliferate and produce a toxin that causes diarrhoea?

00:24:35

(A) Enterococcus species

(B) S. epidermidis

(C) Pseudomonas aeruginosa

(D) Clostridium difficile

A. (D) Clostridium difficil

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Chapter 23

BONE & JOINT INFECTION

OSTEOMYELITIS

Osteo → Bone

Myelitis → Bone Marrow → Inflammation

Types

- Acute osteomyelitis
- Chronic osteomyelitis

Acute Osteomyelitis 00:03:46

- Mainly happens after
  - i) Skin infection (boils) gets hematogenous access
  - ii) After Trauma – Direct contamination of bone

  MC Route → Hematogenous route
- Abrupt in onset
- Progress rapidly
- MCC of acute osteomyelitis – Staph Aureus

Chronic Osteomyelitis 00:05:44

Predisposing condition → Diabetes mellitus

C/F 00:08:40

- Bone pain & tenderness
- Fever, night sweats, fatigue
- Vertebral osteomyelitis
  - Lumber (60%) > Cranial or Thoracic
  - Back pain

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PrepLadder

- Indolent
- Necrosis of bone occurs causing formation of sequestrum (An avascular piece of Infected Bone)

Pathogens

00:10:28

Predisposing Factor	Common Organisms
Neonates	S. agalactiae (group B Streptococcus)
Children and adults	S. aureus
Adults with vertebral osteomyelitis	S. aureus, MTB
Intravenous drug users	S. aureus, P. aeruginosa, Serratia marcescens, Candida albicans
Puncture wounds of foot	P. aeruginosa
Cat bite	Pasteurella multocida
Sickle cell anemia	Salmonella species
Exposure in endemic area	Coccidioides immitis, Histoplasma capsulatum

Treatment

00:16:59

Emperial Rx against S. aureus

1. Vancomycin (most of organisms are MRSA)
2. Naficillin or other cephalosporins given Parentrally

In cases:

Vancomycin Intermediate Sensitive S. aureus (VISA) } Daptomycin/ Linezolid
Vancomycin Resistant S. aureus (VRSA) } Daptomycin/ Linezolid

- Surgical debridement must be done in cases of Chronic Osteomyelitis

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INFECTIOUS / SEPTIC ARTHRITIS

00:19:42

- Main organism involved → S. Aureus
- Monoarticular involvement of large weight bearing joint like hip / knee joint

Analysis of Synovial Fluid for Diagnosis

Disease	Appearance	Cell Number (per μL)	Glucose (Fluid / blood ratio)
Normal	Clear	<200 neutrophils	Approx 1.0
Infectious (septic)	Cloudy	>20,000 neutrophils	<0.25
Rheumatoid arthritis	Opalescent	2000-20,000 neutrophils	0.5-0.8
Trauma	Clear	200-2000 neutrophils	1.0

Clinical Manifestations:

00:23:36

- Acute onset of inflamed joint (Hip or knee joint)
- Fever

On physical examination:

- Joint is Red, warm, swelling along with joint effusion
- In children there is reluctance in moving the joint → Joint immobility d/t pain

Pathogens

00:25:28

- MCC → S. aureus
- In young sexually active individual with multiple partners person develops joint pain → Gonococci
- In patient with hip & knee Prosthesis → S. aureus
- IV drug abusers → S. aureus & Pseudomonas Aeruginosa
- Borrelia Burgodorferi → Lyme's Disease (Spirochaete transmitted via Tick)

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- Erythema chronica migrans Bullous lesion

- Arthritis in Lyme's disease is immune mediated therefore isolation of Bacteria is not possible

Diagnosis

00:30:38

- Gram Stain of Joint fluid
- Blood cultures are positive only in <30% cases
  - 10ml blood with 100ml Glucose Booth
- Radiological features-MRI
  - Soft tissue swellings
  - Destruction of Joints occur & Reduced mobility

Treatment

- Superficial Rx for Staph. Aureus

  i) Vancomycin

  ii) Nafacillin

- DOC is Ceftriaxone for Gonococci infection
- Remove Joint fluid through Arthrocentesis & provide culture specific Antibiotic

VIRAL ARTHRITIS (Immune Complex Mediated)

00:34:17

Immune complex gets deposited on Joints causing Arthritis / Arthralgia inducing inflammatory response

C/F

- Arthralgia – Painful Joint without visible inflammation
- Short duration which goes off with time
- Small Joints of U/L involvement
- Rubella in its course of infection can cause viral arthritis
- Parvo virus B_{19} a/w Arthralgia mimics RA
- Arthralgia & Arthritis also a/w prodromal period of Hep B
- Hep C infection also a/w arthralgia
- Dengue & Chickengunia a/w severe Arthralgia (Arbo virus)

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REACTIVE ARTHRITIS

00:43:57

After GI Infection / Genito Urinary Infection

↓

Develops Joint pain

- It is d/t immune response to bacterial infection
- More seen in HLAB27 positive individuals as they are more prone to reactive arthritis
- Organisms a/w Reactive arthritis
  - Campylobacter
  - Chlamydia
  - Yersenia
  - Shigella
  - Salmonella

C/F

- Assymetric Arthritis of knee & ankle and accompanied by fever
- Resolves within days / weeks
- Recurrence are common
- Culture of synovial fluid always negative

REITER SYNDROME

- Conjunctivitis
- Urethritis
- Reactive Arthritis

Treatment

- NSAIDS (mainstay)
- Antibiotics have no role

RHEUMATIC FEVER

00:49:48

- Immune mediated post streptococcal disease (streptococcus pyogenes) → Group A

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Affects:

- Joints
- Heart
- Brain
- Skin
- Age: 5-15yr
- It begins with migratory Polyarthritis involving the large joints after pharyngitis
- Carditis often occurs in this case & mainly Pancarditis which results in CHF
- Mitral valve →MC involved
- Chorea →Involuntary movements
- Skin →Erythema marginatum & subcutaneous nodule

Jones criteria

00:55:26

A. For all Patient Populations with Evidence of preceding Group A Streptococcal Infection	
Diagnosis: initial ARF	2 major or 1 major manifestations + 2 minor manifestations
Diagnosis: recurrent ARF	2 major or 1 major & 2 minor or 3 minor
B. Major Criteria	
Low risk populations	Moderate- and high risk populations
Carditis	Carditis
• Clinical and / or subclinical	• Clinical and / or subclinical
Arthritis	Arthritis
• Polyarthritis only	• Monoarthritis or polyarthritis • Polyarthragia
Chorea	Chorea
Erythema marginatum	Erythema marginatum
SC nodules	SC nodules

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C. Minor criteria	
Low risk populations	Moderate and high risk populations
Polyarthralgia	Monoarthralgia
Fever (≥38.5°c)	Fever (≥38°c)
ESR ≥60mm in the first hour and / or CRP ≥3.0mg/dL	ESR ≥30mm/hr and / or CRP ≥3.0mg/dL
Prolonged PR interval, after accounting for age variability (unless carditis is a major criterion)	Prolonged PR interval, after accounting for age variability (unless carditis is a major criterion)

Low risk

1. <1/1000
2. School children <2 /1 lakh

In addition lab evidence of prior infection by S. pyogenes is needed it consist of

i) Positive throat culture or positive rapid streptococcal Ag test
ii) Rising streptolysin O Ab Titer
   - ASO >200 todd units

Treatment

- DOC → Aspirin to reduce infection
- Antibiotics → Penicillin G → have no effect on cause of disease but can be given to reduce carriage of streptococci in pharynx
- Prevention of RF involves prompt diagnosis & treatment of Streptococcus throat with penicillin G or oral penicillin V
- In patient with residual heart disease, prevention of additional damage to heart valves by preventing subsequent episodes of streptococcal pharyngitis is very important
- This is achieved by monthly administration of Benzathine penicillin G, a depot preparation
- This should continue until the patient is atleast 20yr old or 10yr after last attack

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Chapter 24

GIT INFECTIONS

INTRODUCTION
00:00:16

- Infections with a variety of agents can occur in any part of the gastrointestinal (GI) tract from the mouth to the anal canal
- Infections can range in severity from self limited to life-threatening, particularly if infection spreads from the gut to other parts of the body
- Infections are typically caused by the ingestion of exogenous pathogens in sufficient quantities to evade host defenses
- And they cause disease by multiplication, toxin production, or invasion through the gastrointestinal mucosa to reach the bloodstream and other tissues.
- In other cases, members of the normal flora of the GI tract can cause disease

ESOPHAGITIS
00:02:17

- Esophagitis is an inflammatory process that can damage the esophagus
- Inflammation caused by infection, typically by fungi such as Candida or viruses such as herpes simplex virus, causes the symptoms of esophagitis.
- Most cases occur in immunocompromised patients, especially those with reduced cell-mediated immunity. The extent of damage to the esophagus is typically related to the severity of symptoms
- Clinical Manifestations Odynophagia and dysphagia are the key clinical manifestations of esophagitis.
- Pathogens – Candida is the MC etiology, particularly HIV infected patients and other immunocompromised hosts.
- Less common pathogens include herpes viruses such as

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PrepLadder

cytomegalovirus and herpes simplex virus

- Noninfectious causes also occur, such as acid reflux from the stomach and medication-induced disease (e.g., doxycycline)
- Diagnosis may be empiric after a trial of fluconazole results in improvement for presumed Candida esophagitis.
- If an empiric course of fluconazole does not work, then endoscopy for visualization and biopsy could be helpful, particulary in immunocompromised hosts.
- Biopsy samples should be analyzed by using pathologic and microbiologic tests

GASTRITIS

00:06:39

- Gastritis refers to inflammation of the mucosa of the stomach
- It may be erosive or non erosive, depending on histologic and endoscopic findings
- A break in the gastric and adjacent duodenal mucosa defines peptic ulcer disease

Pathophysiology

- The mechanism by which one of the main pathogens, Helicobacter pylori causes peptic ulcer disease has been largely elucidated. Following attachment to the gastric mucosa
- H. pylori causes direct mucosal damage by the combination of ammonia production (from the action of the organism's urease on urea) and the host inflammatory response

Clinical Manifestations of Gastritis

00:08:39

- Patients with gastritis typically complain of dyspepsia (epigastric pain, burning), nausea, and vomiting. In the case of peptic ulcer disease, epigastric pain is the primary symptom
- Some patients may report alleviation of pain with food
- Gastrointestinal bleeding is a complication of peptic ulcer disease. Some patients with gastritis may be asymptomatic.

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Pathogens

- Infectious and noninfectious etiologies are possible.

Among infectious causes, H. pylori is the most important

- Viruses such as cytomegalovirus and fungi such as Mucor may rarely cause ulcer disease
- Following ingestion of raw fish, larvae of Anisakis species
- Mycobacteria (tuberculosis and non tuberculosis mycobacterial), Giardia, and Strongyloides may also cause gastritis
- Noninfectious causes such as alcohol and medications (NSAID'S)

Diagnosis

- Upper endoscopy with gastric biopsy is the definitive diagnostic strategy.
- H. pylori-associated ulcers can be confirmed by a urease test on the biopsy specimen
- Noninvasive tests such as the urea breath test or stool antigen test

Treatment

- Directed at the underlying pathogen, taking the host immune status into consideration.
- For H. pylori, combination therapy with two antibiotics.
  Ampicillin and Clarithromycin, + a proton pump inhibitor (omeperazole, or Bismuth)

DIARRHEA

00:13:43

- Acute diarrhea classified as non inflammatory (watery, Non bloody) or inflammatory (bloody).
- Bloody diarrhea is also known as dysentery.
  For example, bloody diarrhea caused by Shigella is often called bacillary dysentery.
- Diarrhea must be calibrated against the patient's normal bowel movements but is usually considered to be greater than three to five bowel movements per day.

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- Most of the infectious agents that cause diarrhea act at the small intestine (where the majority of fluid normally gets absorbed) or the colon

Infectious Agents of Acute Diarrhea and the Underlying Mechanism

Non-inflammatory	Location: Proximal small bowel Illnesss: Watery diarrhea Stool findings: No fecal leukocytes Fecal lactoferrin-not increased	Bacteria: (Mostly Enterotoxin mediated) • Vibrio cholerae • Escherichia coli: • Enteropathogenic • Enterotoxigenic • Enteroaggregative • Clostridium perfringens • Bacillus cereus • Staphylococcus aureus • Staphylococcus aureus • Aeromonas hydrophila • Plesiomonas shigelloides	Viruses: • Rotavirus • Norovirus • Enteric adenoviruses Parasites: • Giardia lamblia • Cryptosporidium species • Cyclospora species • Cystoisospora species • Microsporidia

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Inflammation (invasion or cytotoxin)	Location: Colon or distal small bowel Illness: • Dysentery of • Inflammatory diarrhea	Predominantly dysentery: • Shigella species • Campylobacter jejuni • Campylobacter E. coli • Enteroinvasive E.coli • Vibrio parahaemolyticus	Parasite (Predominantly dysentery) • Entamoeba histolytica • Balantidium coli
	Stool findings: • Fecal pus cells (Polymorphonuclear leukocytes) - increased • Fecal lactoferrin -increased	Predominantly inflammatory diarrhea • Salmonella species • Yersinia enterocolitica • Listeria monocytogenes • Clostridium difficile • Aeromonas hydrophila plesiomonas shigelloideas	
Penetrating	Location: Distal small bowel Illness: Enteric fever Stool findings: Fecal mononuclear leukocytes (↑)	• Salmonella typhi (enteric fever) • Yersinia enterocolitica (typhoid like illness)	

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PrepLadder

Infectious Agents of Food Poisoning

00:23:53

Incubation period,	Symptoms	Common food sources
1-6 hr		
S. aureus	Nausea, vomiting, diarrhea	Ham, poultry, potato or egg salad, mayonnaise, pastries
Bacillus cereus	Nausea, vomiting, diarrhea	Fried rice
Clostridium botulinum	Nausea, vomiting, constipation	Canned food
8-16 hr		
Clostridium perfringens	Abdominal cramps, diarrhea (vomiting rare)	Beef, poultry, legumes, gravies
B. cereus	Abdominal cramps, diarrhea (vomiting rare)	Meats, vegetables, dried beans, cereals
› 16 hr		
Vibrio cholerae	Watery diarrhea	Shellfish, water
Enterotoxigenic E.coli	Watery diarrhea	Salads, cheese, meat water
Enterohemorrhagic E.coli	Bloody diarrhea	Ground beef, salami, raw milk, raw vegetables, apple juice
Salmonella species	Inflammatory diarrhea	Beef, poultry, eggs, dairy products
Campylobacter jejuni	Inflammatory diarrhea	Poultry, raw milk
Shigella species	Dysentery	Potato or egg salad, lettuce, raw vegetables
Vibrio parahaemolyticus	Dysentery	Mollusks, crustaceans

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PrepLa

Agents Causing Traveler's Diarrhea

ViP ECG SCANER

- Vi Vibrio cholera
- P Plesiomonas
- E Entamoeba
- C Cryptospridium
- G Giardia
- S Salmonella & Shigella
- C Campylobacter
- A Aeromonas
- N Norovirus → a/w cruise ships
- E ETEC, EAEC (most imp. cause)
- R Rotavirus

Identification Features/ Detection Methods of Common Organisms Causing Acute Diarrhea or Dysentery

	Presentation	Identification features
Vibrio cholerae	Watery diarrhea	• Darting motility • Coma-shaped Gram -negative bacilli in culture smear • Catalase and oxidase positive • TCBS agar: sucrose fermenting yellow colored colonies • ICUT tests: I+C+U-TSI • Agglutinates with Vibrio cholerae 01 antisera and Ogawa antisera
Shigella	Dysentery	• Gram -ve bacilli, non-motile • Catalase +ve oxidase -ve • MAC or DCA: NLF translucent • XLD: red colonies without black center • ICUT test: I- C+ U- TSI • Agglutinates with Shigella polyvalent antisera and specific monovalent antisera

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PrepLadder

Group B Salmonella	Inflammatory diarrhea	• Gram –ve bacilli, motile • Catalase positive, oxidase –ve • MAC: non-lactose fermenting translucent colonies • DCA: non-lactose fermenting colonies with black center • XLD: red colonies with black center • ICUT tests: I- C+ U- TSI (alkaline slant/ acidic butt, gas, H_2S) • Agglutinates with Salmonella poly-O antisera and serotype (O_4) specific

APPENDICITIS

00:45:05

- Appendicitis is inflammation of the vestigial vermiform appendix. It is one of the most common causes of acute abdomen requiring surgical exploration.

Pathophysiology

- Obstruction of the appendix by one of a variety of causes (e.g., fecoliths, infection such as parasites, tumor) leads to an increase in luminal and intramural pressure.

Clinical Manifestations

- Initial Symptom → Indiagestion
- abdominal pain (especially periumbilical migrating to right lower quadrant) anorexia, nausea, and vomiting.
- abdominal CT scan with contrast is often used when appendicitis is suspected.
- Yersinia, Campylobacter, and Salmonella can cause an acute ileitis and mesenteric adenitis that can mimic appendicitis

Diagnosis

- Clinical manifestations combined with imaging
- Treatment → Surgery is the definitive treatment for appendicitis, usually in concert with perioperative antibiotics.

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DIVERTICULITIS

00:49:29

- Diverticulitis is inflammation of a sac-like protrusion of the colonic wall, usually in the sigmoid colon.

- Perforation of the diverticulum with consequent abscess formation or peritonitis may occur

Pathophysiology

- Colonic diverticula may occur following years of a diet deficient in fiber.

Clinical Manifestations

- There is a range of symptoms depending on the degree of perforation.

- Patients usually present with dull, aching, left lower quadrant abdominal Pain. This is often accompanied by a low-grade fever, leukocytosis nausea, and vomiting. Diarrhea or constipation may be present.

- If perforation occurs, patients can present with generalized peritoneal signs with diffuse abdominal pain and shock.

Pathogens

- Bowel flora such as anaerobes, i.e B fragilis and those in the Enterobacteriaceae family, such as E. coli, are typically involved.

Diagnosis

- Abdominal CT will show colonic diverticula and associated wall thickening, fat infiltration, abscesses, and extraluminal air or contrast medium

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ENTERIC FEVER

00:52:59

- Enteric fever is a clinical syndrome comprised of constitutional symptoms, such as fever, headache, nausea, vomiting and abdominal pain caused by the Salmonella typhi.

- Following the consumption of contaminated food. Salmonella bacteria enter through the intestinal mucosal epithelium by transcytosis.

- The microbes then replicate in the macrophages of Peyer patches, mesenteric lymph nodes, and the spleen.

- Bacteremia then occurs with dissemination to lungs, gallbladder, kidneys, or central nervous system.

- Humans are the only reservoir for S. typhi, so contamination of food or water by human feces should be suspected

Clinical Manifestations

- A prodromal phase is characterized by constitutional symptoms such as malaise, together with abdominal pain, constipation and headache.

- Fever increases over the next several days. Diarrhea is uncommon

- During the second week of disease, a typical transient rash of pink maculopapular lesions (rose spots) may be seen.

- Spleenomegaly occurs more commonly than hepatomegaly, but both may occur. Relative bradycardia and leukopenia are often observed

- The chronic carrier state occurs in approximately 3% of patients with typhoid fever.

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- The organisms typically reside in the gallbladder and are excreted in the stool, serving as a source of infection for others.

Pathogens

- S. typhi and other Salmonella species, such as Salmonella paratyphi A and S. paratyphi B. cause Enteric fever

Diagnosis

- A history of travel to endemic areas, together with a compatible clinical presentation, is often used initially.
- Any fever in a returning traveler should prompt blood culture and a clinical suspicion for enteric fever.

1st week	B	Blood culture/ Bone marrow culture
2nd week	A	Agglutination
3rd week	S	Stool culture
4th week	U	Urine culture

- Early in the disease, blood cultures are typically positive and stool cultures are often negative.

- Later in the disease and in the carrier state, stool cultures are positive and blood cultures are negative.

- Stool cultures are positive at this stage because bile from an infected gallbladder carries organisms into the stool

Treatment

00:59:25

- Intravenous Ceftriaxone is effective treatment modality.
- Cholecystectomy should be considered for those chronic carriers who do not respond to antimicrobial therapy.

- Prevention Hygienic measures to protect the food and water supply from Human fecal contamination are an important public health intervention.

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- Immunization may not always be effective but can be considered in epidemic outbreaks, for travelers to endemic countries and for household contacts of typhoid carriers.

Vaccine

- Two vaccines against typhoid fever are available in the United States, both providing approximately 50% to 80% protection.

- The vaccine containing the Vi capsular polysaccharide of S. typhi has the advantage of being administered once IM

- The other vaccine contains live attenuated S. typhi organisms and is administered orally. It has the advantage of stimulating gut immunity thereby interrupting transmission

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Food Poisoning

Incubation Period > 16 hours

1) Vibrio cholerae – Rice watery stools
   - Shellfish
2) Enterotoxigenic E. coli – Salad, cheese
3) Enterohaemorrhagic E. coli – Ground beef, salami, raw milk
4) Salmonella – Poultry, eggs
5) Campylobacter – Poultry, raw milk
6) Shigella - Potato or egg salad, lettuce.
7) Vibrio parahaemolyticus: - Oyster

Incubation Period: 1-6 hours

1) S. aureus : Ham, poultry, pastries, mayonnaise, potato or egg salad
2) B. cereus (Emetic) : Fried rice/ Chinese Food
3) Cl. Botulinum : Canned food

Incubation Period : 8-16 hours

1) Cl. Perfringens → Beef, poultry, legumes, gravis
2) B. cereus (diarrhea) : Meat, vegetables.

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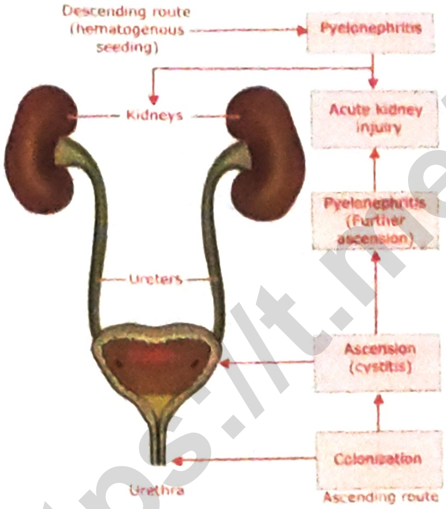
Chapter 25

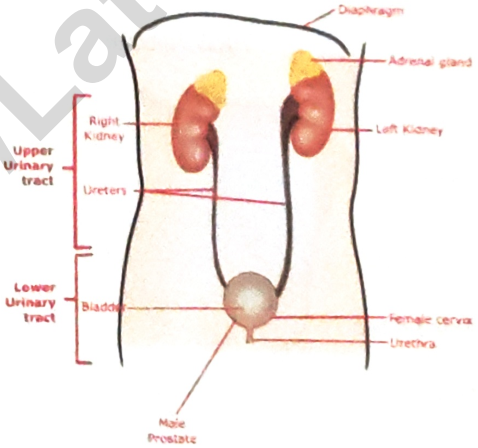
URINARY TRACT INFECTIONS

Disease of Urinary Tract in which there is microbial invasion of Urinary Tract

00:01:31

	Lower UTI	Upper UTI
Site	Urethra & Bladder	Kidney & Ureter
Symptoms	Dysuria, Urgency, Frequency	Fever, Vomiting Abdominal/ flank pain
Route of spread	Ascending	Can be Ascending or Descending





Predisposing Factors of UTI

00:06:55

1) Gender → F > M
   - Short urethra
   - Close proximity of urethral meatus to anus
2) Age → Incidence increase with age
3) Pregnancy

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- Anatomical & Hormonal changes presdispose to UTI
- Asymptomatic, Bacteriurea needs to be treated

4) Structural & Functional Abnormality

Obstruction	Neurogenic Bladder d/t:
– Renal & uretric stone	– Spinal cord injury
– Prostate in males	– Multiple sclerosis
– Any tumors	

5) Virulence of Bacteria → Express pili or fimbriae which helps in adhesion to uroepithelium
   → P. fimbriae are mannose resistant & leads to more destruction

6) Vesico Uretric Reflex (VUR)
   - Valve mechanism is defective
   - Allow urine from bladder into ureter allowing Retrograde movement

7) Genetic factors

Etiology 00:17:50

1) Uropathogenic E. coli → MCC of UTI
   MC nosocommial infection → UTI
2) Enterobacteriacae family
3) Candida albicans
4) Certain staph → Staph saprophyticus → UTI in young female associated with Honeymoon Cystitis
5) Enterococcus
6) Parasites → T. vaginalis, Schistosoma Hematobium
7) Viruses → HSV
   Adeno virus
   CMV

Descending route (Invasion of renal Parenchyma) & Hematogenous spread, occurs d/t Bacteria

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Cause by S. aureus
Salmonella
MTB
Leptospira
Candida

CYSTITIS

00:23:56

Inflammation of Bladder

C/F → Dysuria, Frequency, urgency
Urine – cloudy
No Hematuria
No associated systemic manifestation

ACUTE URETHRAL SYNDROME

00:25:36

Lower UTI

Mainly in young sexually active female
Bacterial count = $10^2 - 10^5$ FU/ml
Pyuria is present

Etiology → Chlamydia, Gonococcus, HSV

Upper UTI

00:27:27

Inflammation of:

- Kidney parenchyma
- Callyces
- Renal pelvis

Systemic Manifestation → Fever, Flank pain, Vomiting
(Absent in Lower UTI)

Lab Diagnosis

00:29:11

Urine specimen Collection

i) MSU Clean catch
ii) Suprapubic aspirate
iii) Cathetrised patient → Taken from cathter tube after clamping

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Transportation: In Urine

i) Add boric acid

ii) Refrigeration

- Direct examination → wet mount examination

↓

Pus cells present

- Leukocyte Esterase Test → Detects esterase enzyme activity
- Nitrate Reduction test → Griess Test

→ Converts Nitrate to nitrite

- Catalase test
- Gram staining

Culture → McConkey agar, CLED (Better) } Differential & Indicator Media

(Since non selective → inhibit growth of proteus)

Get colony count

↓

Multiply with dilution factor (10^3)

↓

If final count is

(i) >10^5 → Significant Bacteriuria

→ Even if patient have no symptom it should be treated for sure

(ii) >10^4 + signs & symptoms of UTI → UTI

(iii) 1 bacteria/ml in Suprapubic Aspirate → UTI

Rx

00:38:54

Norfloxaicn

Nitrofurantoin

Antibiotic sensitive testing should be done on MHA by Kirby Baur Disc Diffusion Test → then prescribe antibiotics

→ Cephalosporin

→ aminoglycosides

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In case of Nosocomial infections (UTI) treat according to the causative organism:

- Piperacillin + Tazobactam
- Fosfomycin
- Carbapenems

00:41:32

Q1. A 30 yr old female with frequency, urgency, dysuria since 2 days

A. Lower UTI

Q2. A 20 yr old female with High grade fever along with vomiting, flank pain & increase in frequency of urination x 3 days

A. Upper UTI

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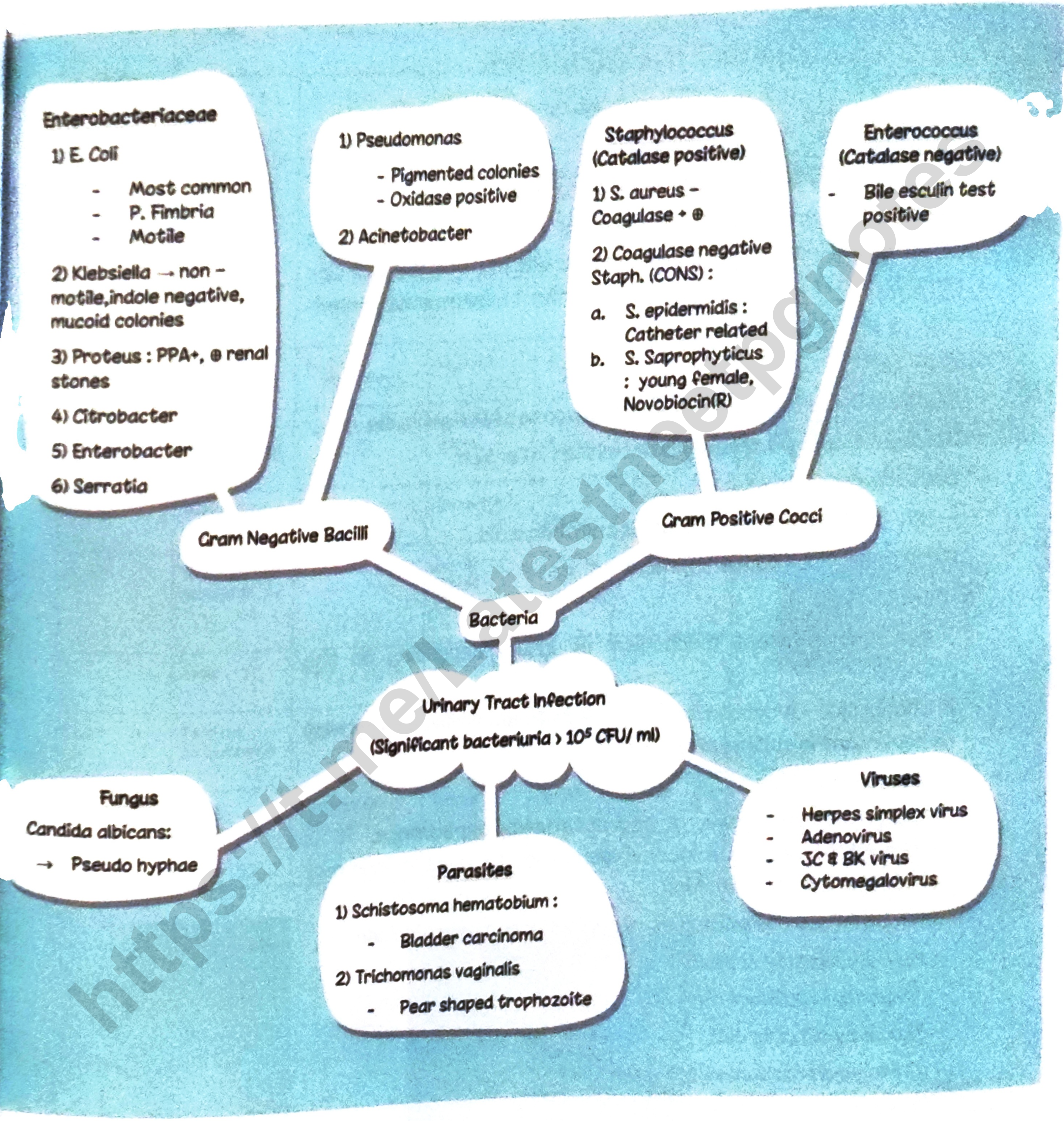

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Chapter 26

Pelvic Infections

Introduction

00:00:13

- Primarily affect sexually active Women & Men
- Most of the pathogens implicated are sexually transmitted, so Partner notification and treatment, as well as patient education regarding safe sexual practices is important step in management

Genital ulcer

00:01:21

- Manifests as a breach in the skin or mucosa of the genitalia
- Usually caused by a sexually transmitted infection.
- Causative organisms
  - HSV-2 (MC) followed by Syphilis and Chancroid
  - Non infectious cause →Behcet's disease

Pathophysiology

- There are different mechanism of injury depending on the pathogen
- In Chancroid → A cytotoxin secreted by Haemophilus ducreyi may be important in epithelial cell injury

Clinical manifestation

- Although lesions have a characteristic appearance, it is important to note the local epidemiology because lesions may appear in an atypical fashion
- Common infections etiologies of genital ulcer disease include
  - HSV-2 (Genital Herpes)
  - Treponema pallidum (Primary Syphilis)
  - H.ducreyi (Chancroid)
  - Chlamydia trachomatis serovars L_1-L_3 (Lymphogranuloma venerum)
- Klebsiella granulomatis (Granuloma inguinale / Donovanosis)

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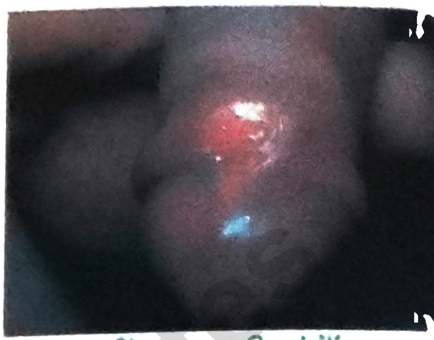
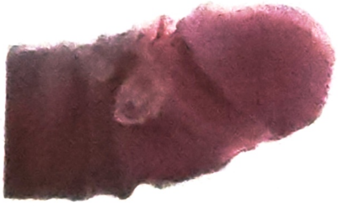
00:04:53

	She Syphilis	Have Herpes	Chronic Chancroid	LGV LGV	Disease Donovanosis
I.P.	9-90 days	2-7 days	1-14 days	3 days - 6 weeks	1-4weeks
Primary lesions	Papule	Vesicle	Pustule	Papule, Pustule, Vesicles	Papule
No. of lesions	1	Multiple	Multiple	1	Variable
Diameter	5-15mm	1-2mm	Variable	2-10mm	Variable
Edges	Sharply demarcated	Erythematous	Undermined edges	Elevate d	Elevated, Irregular
Depth	Superficial or deep	Superficial	Excavated	Sup. / deep	Elevated
Base	Smooth, non purulent	Serous red	Purulent, bleeds easily	Variable , non vascular	Red velvety
Indurations	Firm	None	Soft	Firm	Firm
Pain	Painless (uncommon)	Tender	Very tender	Variable	Uncommon
LAP	Firm, nontender B/L	Firm tender B/L	Tender	Tender	Pseudo-buboes (nontender)


Chancre Syphilis

Genital Herpes


LGV

Chancroid

Donovanosis

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Diagnosis

- Thorough sexual and medical history f/b physical examination
- Clinical characteristics are very helpful but there can be overlap in presentation, and there may also be multiple syndromes co-presenting
- Diagnostic testing is highly recommended, testing for other sexually transmitted diseases including HIV is also important because there is often co-transmission of multiple pathogens

Treatment

- DOC for Genital Herpes→ Acyclovir or one of its derivatives, Famiciclovir or Valacyclovir
- Primary and secondary syphilis → Long acting penicillin (Benzathine penicillin G)
- DOC for Chancroid →Azythromycin
- DOC for Lymphagranuloma venerum →Doxycycline

Prevention strategies

- Consistent use of condoms is an important measure that can prevent genital ulcers
- Partner notification & treatment are important prevention strategies as well

VAGINITIS 00:29:30

- Inflammation of Vagina that can result in discharge, itching and pain
- Common causes of vaginitis are
  - Candidiasis
  - Trichomoniasis
  - Bacterial vaginosis
  - Non infectious causes
    - Lichen Planus
    - Certain medications eg. Oral contraceptives

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Pathophysiology

- Use of Antibiotics that inhibit the normal flora of the vagina (especially lactobacilli), prediposes to candida vaginitis. Candida is a member of Normal flora of many women
- Bacterial vaginosis →Not a sexually transmitted disease.
- Trichomoniasis→Sexually transmitted disease
- White "cottage cheese" appearance →Vaginal candidasis
- Strawberry cervix →Trichomoniasis.
- Bacterial vaginosis → Discharge is thin, grayish with unpleasant odor, often described as Fishy (Whiff test +ve)

Feature	Normal findings	Candidiasis	Trichomoniasis	Bacterial vaginosis
Symptoms	Nothing	Vulval itching	Produce discharge, itching	Bad odour, increased discharge
Discharge feature	Variable / scanty / clear	Scanty white clumps	Profuse, white / yellow or green coloured discharge	White / grey
Inflammation of vaginal epithelium	None	Redness of vaginal epithelium Vulvar dermatitis	Redness of vaginal epithelium Colpitis macularis (Strawberry cervix)	None
PH*	Normal <4.5	<4.5	>5	>4.5 (Amsels criteria)
Microscopy	N. epithelial cells, lactobacilli	Leucocytes, pseudo-hyphae	Leucocytes, Trichomonas in Urine Microscopy	Clue cells (Nugent score)
	NAAT (Nucleic Acid Amplification Test)			
Treatment		Clotrimazole 100mg vaginal tab OD x 7days or Fluconazole 150mg orally (single dose)	Metronidazole 500mg BD x 7 days (orally)	

Candida Vulvovaginitis

Trichomoniasis

Clue cells

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CERVICITIS

00:50:43

Cervicitis

- Cervicitis → Inflammation of the Uterine cervix.
- Acute cervicitis → Usually due to a sexually transmitted infection caused by either C. trachomatis or Neisseria gonorrhoeae (MC) or both

Clinical Manifestations

- Asymptomatic in most of cases.
- Detected on speculum examination and following routine screening to C.trachomatis and N. gonorrhoeae
- Women with concomitant urethral infection may have dysuria
- Physical examination → Friability of the cervical tissue
- Pathogens → C. trachomatis (Serovars D-K), N. gonorrhoae, HSV and T.vaginalis
- Diagnosis
  - Clinical diagnosis → Increased friability of the cervix, with or without mucopurulent discharge.
  - Laboratory diagnosis
    - NAAT for C. trachomatis and N. gonorrhoeae.
    - Gram stain, Culture (If NAAT testing not available)

- Treatment (for both C. trachomatis and N. gonorrhoeae)
  - Ceftriaxone intramuscularly + Azithomycin orally
  - Treat the partner too
- Prevention
  - Consistent use of Condoms
  - Partner notification and treatment

PELVIC INFLAMMATORY DISEASE

00:54:36

- Polymicrobial infection of upper genital tract structures (Uterus, Fallopian tubes and Ovaries)
- Endocervical canal barrier compromised → Vaginal bacteria ascend into normally sterile space of the upper genital tract
- STD's affecting the Cervix can initiate the process, permitting the anaerobic bacteria of the vagina to ascend

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- Having multiple sex partner increase the risk of PID
- Multiple episodes of PID lead to scarring of the fallopian tubes and an increased risk of ectopic pregnancy and sterility

Clinical Manifestation

- Symptoms → Lower back pain, fever, chills, lower abdominal pain, Cervical and adnexal tenderness
- Pathogens → N. gonorrhoeae, C. trachomatis alongwith enteric gram-negative rods and anaerobes
- Diagnosis
  - Uterine, adnexal, or cervical motion tenderness
  - Fever, presence of leukocytes on cervical / vaginal discharge, elevated C-reactive protein
  - Laboratory evidence of cervical infection with N. gonorrhoeae or C. trachomatis

Treatment

- Mild disease
  - Cefoxitin / Ceftriaxone (one dose) plus doxycycline (14 days) ± Metronidazole (OPD based treatment)
- Moderate to severe disease
  - IV Cefoxitin / Cefotetan + Doxycyciline or Clindamycin + gentamicin (IPD based treatment)
  - This is f/b Oral antibiotics only after 24 hours of improvement of the patient

Urethritis

01:02:02

- Definition → Inflammation of the Urethra, usually caused by STD, particularly in sexually active men.
- Noninfectious causes → Reiter's syndrome (Urethritis, Uveitis, and Reactive arthritis)
- Clinical manifestations- Dysuria, discharge from Urethra, Pruritis, burning sensation

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- Pathogens
  - N. gonorrhoeae and C. trachomatis (MC), Mycoplasma genitalium and T. vaginalis.
- Diagnosis - NAAT for C trachomatis and N. gonorrhoeae

Prostatitis

01:03:48

- Acute bacterial prostatitis
  - Characterized by typical irritative voiding symptoms urinary frequency, hesitancy, feeling of incomplete voiding, dribbling, fever, pyuria, and positive urine cultures
- Chronic bacterial prostatitis
  - Characterized by the same voiding symptoms, but fever and pyuria are typically absent
- Pathophysiology
  - Bacteria ascend the urethra, and then reflux into the prostatic ducts where infection occurs
- Clinical manifestations
- Acute Prostatitis
  - Patients appear ill with fever, chills, irritative voiding symptoms, and pelvic or perineal pain.
  - Physical examination reveals a very tender and enlarged prostate
- Chronic Prostatitis
  - Symptoms are more subtle.
  - Patients may present with Recurrent UTI
  - Prolonged treatment of prostatitis cures the disease

- Pathogens
- Gram negative rods, Enterobactericeae (eg. Escherichia coli, Kiebsiella and Proteus species), Pseudomonas
- N. gonorrhoeae and C trachomatic cause prostatitis, in a/w urethiritis and epididymitis in sexually active men

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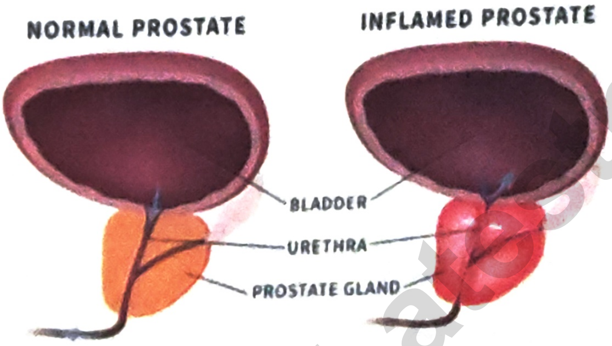
- Diagnosis
  - Edematous and tender prostate
  - Urine culture →Done to determine causative organisms
  - Prostatic fluid culture →Not done in Acute Prostatitis because prostatic massage should not be done during the acute phase
  - Prostatic massage may be useful in Chronic Prostatitis
- Treatment
  - Trimethoprim-Sulfamethoxazole or Ciprofloxacin can be used as empiric therapy until culture results return
- These agents exhibit good penetration into the prostate. Therapy is prolonged (4 to 6 weeks)



Prostatitis

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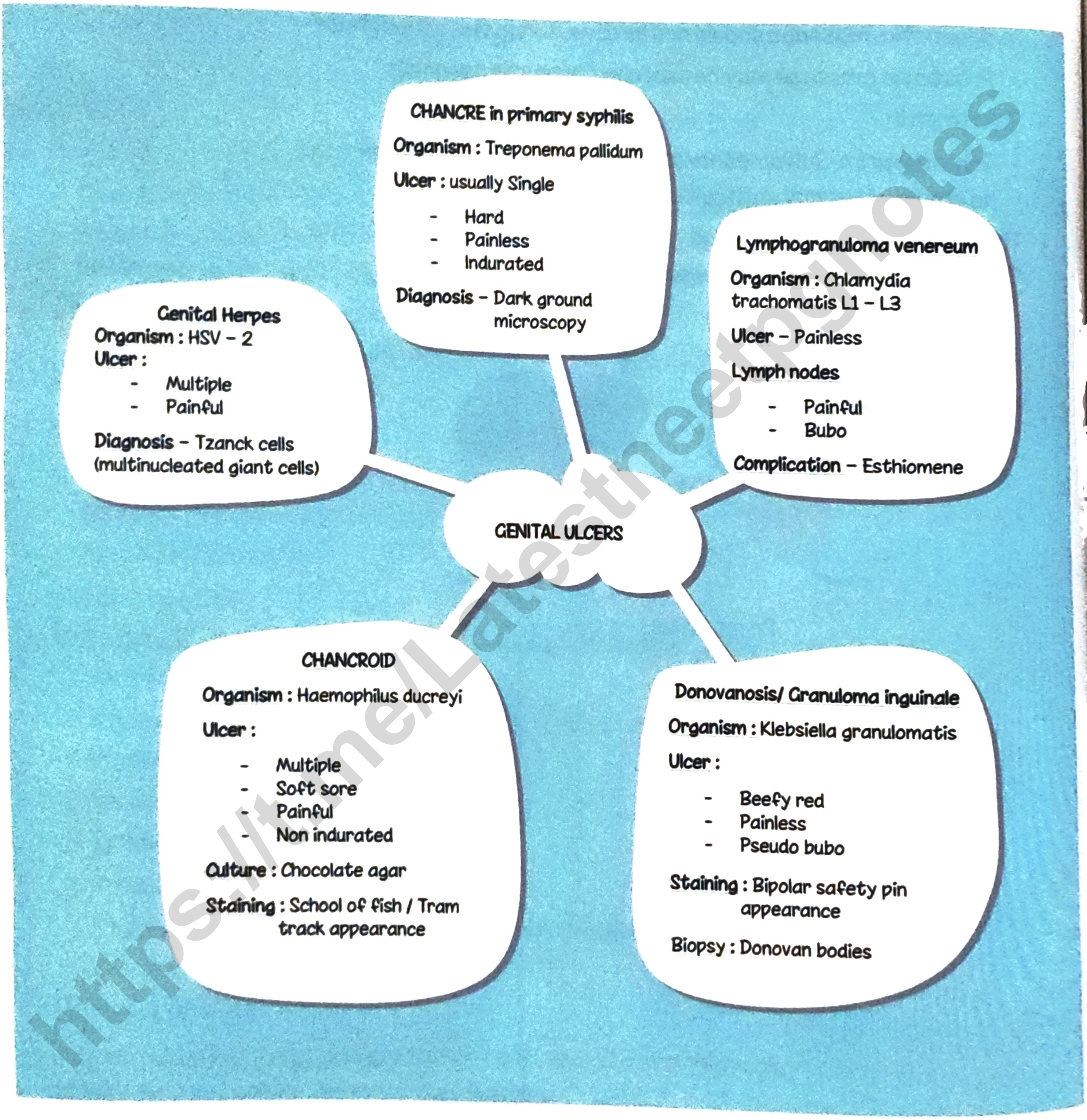

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Chapter 27

CARDIAC INFECTIONS

INTRODUCTION

00:00:26

- Cardiac infections are severe, life threatening infections in many cases. The heart valves endocardium, myocardium, and pericardium can all be infected

- In addition, infection of cardiac devices (pacemakers, defibrillators) is becoming more frequently diagnosed with their increase in use.

ENDOCARDITIS

00:01:17

- Endocarditis is an infection of the valves of the heart.

Pathophysiology

- Infection of the heart valves is thought to result from the colonization of damaged valvular endothelium by circulating pathogens.

- Endothelial damage may result from turbulent blood flow around the valve (congenital or rheumatic heart disease), direct injury from foreign bodies (e.g., intravenous catheters), or repeated intravenous injections of particles in intravenous drug users.

- Organisms enter the bloodstream most often at the site of dental surgery, indwelling intravenous catheters, or intravenous drug use.

- Adhesion of bacteria to the damaged endothelium is enhanced by their ability to produce a glycocalyx

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- Once the infection has begun, a combination of organisms and thrombus organize to form a vegetation

- Destruction of the valve occurs at different rates depending on the virulence of the organism. As the valve is destroyed, symptoms of valvular regurgitation can develop.

- Organisms can spread to surrounding myocardium. resulting in abscess formation and destruction of the electrical conduction system

- As the vegetation on the valve enlarges, fragments can spread via the blood stream (emboli), resulting in catastrophic effects, such as CVA and metastatic infections

Organism(s)	Percentage of cases								
	Native Valve Endocarditis		Prosthetic Valve Endocarditis at indicated time of onset (months) after valve surgery			Endocarditis in IV drug users			
	Community acquired	Health care associated	< 2	2-12	> 12	Right sided	Left sided	Total	CED
Streptococci	40	13	1	9	31	5	15	12	2
Pneumococci	2	-	-	-	-	-	-	-	-
Enterococci	9	16	8	12	11	2	24	9	4
Staphylococcus aureus	28	52	22	12	18	77	23	57	36
Coagulase -ve staphylococci	5	11	31	32	11	-	-	-	41
Fastidious gram -ve cocco bacilli (HACEK group)	3	-	-	-	6	-	-	-	-
Gram -ve bacilli	1	1	13	3	6	5	13	7	6
Candida spp.	<1	1	8	12	1	-	12	4	2
Polymicrobial	3	3	3	6	5	8	10	7	2
Diphtheroids	-	<1	6	-	3	-	-	0.1	1
Culture -ve	9	3	5	6	8	3	3	3	6

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HACEK Organisms

- Haemophilus aphrophilus and Haemophilus paraphrophilus, now called Aggregatibacter aphrophilus and Aggregatibacter paraaphrophilus
- Actinobacillus actinomycetemcomitans, now called Aggregatibacter actinomycetemcomitans
- Cardiobacterium hominis
- Eikenella corrodens and
- Kingella kingae

DOC: Ceftriaxone

- Pathogens that do not grow in routine blood culture media and require specialized testing for diagnosis include:
  - Bartonella species, Coxiella burnetii, Brucella species, and Tropheryma whipplei. These are cited as pathogens that cause "culture-negative" endocarditis.
  - The most frequent cause of "culture-negative endocarditis" is the use of antimicrobials prior to obtaining blood cultures

Clinical Manifestations

- Depending on the virulence of the infecting pathogen. the time course of illness may be days
- Acute endocarditis
  - Caused by Staphylococcus aureus
- Subacute endocarditis
  - Caused by viridans group streptococci
- Constitutional symptoms: fever (>80% cases). Chills, night sweats,

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anorexia

- Consequences of destruction of heart valves and associated structures: new murmur, heart failure, atrioventricular (AV) block (PR prolongation seen on ECG)

- Embolic phenomenon: Left-sided endocarditis: CVAs or brain abscess (new focal neurologic deficits), spleenic or renal infarcts (abdominal or flank pain), and emboli to others sites manifesting as splinter hemorrhages , Janeway lesions , retinal hemorrhages, and conjunctival hemorrhages

- Right-sided endocarditis: septic pulmonary emboli (cough, shortness of breath, chest pain, hemoptysis)

- Antigen-antibody deposition from uncontrolled infection: Osler's nodes, Roth's spots, glomerulonephritis (hematuria), and/or arthritis

Roth's spots (Litten's Sign)

Janeway Lesion

Osler's nodes (Painful, red, raised lesion)

Splinter Haemorrage

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Clinical and Laboratory Features of Infective Endocarditis

Frequency	Frequency (%)
Fever	80-90
Chills and sweats	40-75
Anorexia, weight loss, malaise	25-50
Myalgias, arthralgias	15-30
Back pain	7-15
Heart murmur	80-85
New/worsened regurgitant murmur	20-50
Arterial emboli	20-50
Splenomegaly	15-50
Clubbing	10-20
Neurologic manifestations	20-40
Peripheral manifestations (Osler's nodes, subungual hemorrhages, Janeway lesions, Roth's spots)	2-15
Petechiae	10-40
Laboratory manifestations	
Anemia	70-90
Leukocytosis	20-30
Microscopic hematuria	30-50
Elevated ESR	60-90
Elevated CRP	>90
Rheumatoid factor	50
Circulating immune complexes	65-100
Decreased serum complement	5-40

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Major criteria

1. Positive blood culture & Any one of the following:
   A. Typical IE organism isolated from two separate sets of blood cultures (Viridans Streptococci, S. gallolyticus, HACEK group, S. aureus or Enterococci) or
   B. Persistently positive blood culture with agents other than typical IE organisms:
   - Blood culture sets drawn > 12 h apart; or
   - All of 3 or majority of > 4 separate sets of blood culture, with first and last drawn at least 1 h apart

   C. Single positive blood culture for Coxiella burnetii or phase 1 IgG antibody titre of > 1:800

2. Evidence of endocardial involvement:
   A. Positive echocardiogram
   - Oscillating intracardiac mass on valve or
   - Abscess or
   - New partial dehiscence of prosthetic valve

   B. New valvular regurgitation

Minor criteria

1. Predisposition: predisposing heart condition or IV drug use
2. Fever > 38.0°C (>100.4° F)
3. Vascular phenomena: major arterial emboli, septic pulmonary infarcts, mycotic aneurysm, intracranial hemorrhage, conjunctival hemorrhages or Janeway lesions
4. Immunologic phenomena: glomerulonephritis. Osler's nodes, Roth's spots or rheumatoid factor
5. Microbiologic evidence: positive blood culture but not meeting major criterion as noted previously or serological evidence of active infection with organism consistent with infective endocarditis

Definite endocarditis if the following are present:

- Two major criteria or

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- One major criterion and three minor criteria or
- Five minor criteria

Endocarditis- Valve Vegetation on Echocardiography

- Evaluation of valves for infection is best accomplished through echocardiography. A transthoracic echocardiogram (TTE) has reduced sensitivity when compared with a transesophageal echocardiogram

- TEE to assess for vegetations and myocardial abscesses but is a less invasive test. Not only can echocardiogram identify new vegetations on valves, which are evidence of infection, but it can also assess the degree of valvular damage and complications such as perivalvular abscesses

- ECG can be used to detect damage to the conducting system. The MC finding is PR prolongation in patients with aortic valve endocarditis and associated perivalvular abscess.

There Are Two Types Of Echocardiograms

- Transthoracic Echocardiogram (TTE), where the probe is placed on the chest wall, and a TEE, where the probe is inserted into the esophagus.

- The TEE often produces higher-quality images, particularly of aortic and mitral valves, since the TEE probe is closer to the heart itself.

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TREATMENT

- Without treatment, endocarditis is always fatal, so prompt effective therapy is essential. Bactericidal drugs should be used.

- antimicrobial therapy
- In some cases, surgical removal of the infected valve
- Empiric therapy for endocarditis: hemodynamic instability, severe disease, evidence of embolic disease, or large vegetations.

- Empiric antimicrobial coverage should be active against MRSA, Viridans group streptococci, Enterococci and HACEK organisms.

- Common empiric regimens include:
  - Vancomycin + Ceftriaxone or Gentamicin
  - Specific antimicrobial therapy should be instituted when the results of blood cultures and antibiotic susceptibility tests are known.
  - Antimicrobial therapy for endocarditis is usually required for 4 to 6 weeks

- Surgical therapy is either indicated or should be strongly considered in patients with severe congestive heart failure perivalvular abscesses, infections refractory to medical management, and embolic events with large vegetations.

Prevention

- In patients with prior endocarditis a prosthetic heart valve, or select types of CHD, antibiotic prophylaxis is recommend prior to certain procedures.

MYOCARDITIS

00:43:40

- Myocarditis is infection of the heart muscle.

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Pathophysiology

- Infection of the myocardium most frequently occurs following hematogenous spread of virus or other pathogen to the heart muscle, although direct spread from adjacent structures can occur. Infection and inflammation of myocardium may result in cardiac dysfunction, leading to heart failure.
- Patients with myocarditis present with signs and symptoms of heart failure.
- Depending on the pathogen, the pace of disease progression may be over days or weeks.
- Patients may have signs and symptoms of a systemic infection as well (fever, constitutional symptoms). Those with associated pericarditis often have chest pain
- Viral pathogens are thought to be the predominant cause of infectious myocarditis, although many cases are idiopathic.
- MCC Coxsackie virus, although CMV, EBV, parvovirus B19, and influenza virus have been implicated. Other pathogens include Trypanosoma cruzi, the agent of Chagas disease and Trichinella spiralis

Diagnosis

- A definitive diagnosis requires cardiac muscle biopsy revealing myocardial inflammation and necrosis.
- However, most cases are presumptively diagnosed in a patient presenting with heart failure, who has (often global) cardiac dysfunction on ECG and elevated cardiac enzymes.
- The ECG may be abnormal and may show ST changes mimicking an acute myocardial infarction

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Treatment

- There is no known treatment for most causes of myocarditis, and supportive care is most often given. Patients may ultimately require heart transplant

PERICARDITIS

00:49:56

- Pericarditis refers to inflammation of the pericardium, which can be due to infection, autoimmune diseases, trauma, or malignancy

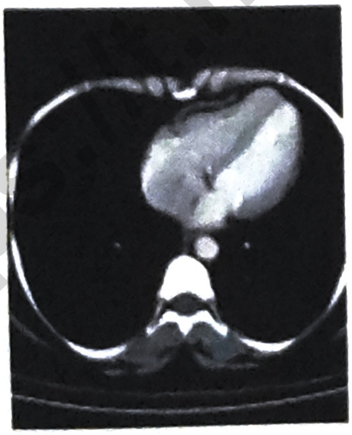
Pathophysiology

- Pathogens reach the pericardium by either hematogenous spread through the blood or direct spread from adjacent intrathoracic structures or, rarely, directly from infected myocardium.

- Inflammation of the pericardium can result in the formation of pericardial effusion. Pericardial effusions can result in cardiac tamponade. Inflammation can also result in a constrictive physiology.

- Certain infections causing pericarditis may also be associated with a concomitant myocarditis

CONSTRICTIVE PERICARDITIS



There is diffuse thickening of the anterior pericardium upto the vascular root

- Chest pain is the MC manifestation of pericarditis. Pain often worsens with inspiration or coughing. Sitting up and leaning forward often improve the pain associated with pericarditis.

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- Patients may have fever and constitutional symptoms.
- On examination a friction rub (often consisting of three phases) may be heard when performing auscultation of the heart. This exam finding is very specific for pericarditis.
- Severe infection may result in cardiac tamponade or constrictive cardiac physiology. These patients present with acute or subacute/chronic onset of symptoms of heart failure, respectively.

Pathophysiology

- Viruses, bacteria, mycobacteria and fungi have all been reported to cause pericarditis.
- Among viral infections: Coxsackie virus and echovirus are MC, although HIV and CMV can cause pericarditis as well.
- Among bacteria: S. aureus and S. Pneumoniae are MC.
- MCC worldwide MTB
- Clinical presentation is often subacute and may result in a constrictive pattern.
- Several fungi such as Histoplasma capsulatum and Coccidioides immitis can cause pericarditis, which clinically presents similarly to tuberculous pericarditis
- An ECG or cardiac magnetic resonance imaging will often reveal a pericardial effusion and/or pericardial thickening.
- In addition, chest X-ray may show an enlarged cardiac silhouette, and cardiac enzymes can be elevated.
- Recovery of a pathogen often requires a pericardiocentesis or

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pericardial biopsy

- Treatment for infectious pericarditis is dependent on the pathogen.

- Most viral etiologies are treated with symptomatic management and supportive care, whereas bacterial. mycobacterial, and fungal infections will require directed antimicrobial therapy.

- In patients with constrictive pericarditis and tamponade, pericardtocentesis can be life-saving. Untreated bacterial pericarditis is rapidly fatal

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Chapter 28

Upper Respiratory Tract Infections

OTITIS MEDIA

00:00:18

- Otitis media is an Infection of the middle ear caused by either viruses or bacteria. Otitis media can be either acute or chronic.

- Any process that leads to eustachian tube obstruction can result in fluid retention and concomitant infection of the middle ear.

- The most common predisposing factors are upper respiratory tract infections and seasonal allergic rhinitis.

- Otitis media is very common in children under the age of 3 years because they have an eustachian tube that is easily blocked by the inflammation caused by a viral infection or an allergic response

Clinical Manifestations

- Patients present with Ear pain and pressure, often accompanied by an upper respiratory tract infection.
- In infants, the ear pain may manifest as ear pulling.

- decreased hearing and fever.
- O/E: The TM is erythematous, loss of the light reflex and decreased mobility.

- In some cases, the tympanic membrane may bulge.

Pathogens

- Both Bacteria and Viruses cause otitis media

- Among Bacteria, Streptococcus pneumonia (MCC)
- Nontypeable strains of Haemophilus influenzae and Moraxella catarrhalis are also common causes.

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- Among viruses, Respiratory syncytial virus Coronaviruses and Rhinoviruses are commonly involved.

Diagnosis

- diagnosed clinically:
  - If the membrane ruptures, a sample of the exudate can be analyzed by Gram stain and culture.

- If indicated, tympanocentesis can be done to relieve pressure before the drum ruptures and to obtain a specimen for culture

Rx

- DOC Amoxicillin orally together with nasal decongestants to open the eustachian tube.

- In cases of bacterial resistance → Amoxicillin-clavulanate (Augmentin) may be used.

- Ventilating tubes may be inserted as a strategy to prevent recurrent infections.

- The conjugate pneumococcal vaccine is effective in preventing invasive pneumococcal disease but is less effective in preventing otitis media

SINUSITIS

00:07:00

- Sinusitis is inflammation of the paranasal sinuses. It can be either acute or chronic.

- Acute Sinusitis: Symptoms < 4 weeks

Pathophysiology –Impaired mucocilliary clearance caused by viral infection or allergic rhinitis can obstruct the orifice of the sinus.

- Mucus then accumulates in the sinus cavity. Stasis can lead to bacterial overgrowth and superinfection.

- Sinusitis frequently involves the maxillary sinus

- Because the ostium of that sinus is located superior to most of

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the sinus and drainage of mucus has to occur against gravity.

- Drainage of the other sinuses is aided by gravity

Clinical manifestations:

- Purulent nasal discharge.
- Nasal congestion
- Facial or anus pain
- Decreased sense of smell and fever.

- Headache and malodorous breath may also be present.

Pathogens

- many cases begin with viral upper respiratory tract infection.
- Bacterial superrinfection can then occur.

- Common organisms are S. pneumoniae. H. influenzae, and M. catarrhalis, as in the case of acute otitis media

- Staphylococcus aureus less common.

- In immunocoporomised patients and diabetics, sinusitis caused by fungi such as Aspergillus or Mucor may occur

Diagnosis

- Based on symptoms and clinical findings.

- CT scan of the sinuses is very sensitive modality for indicating inflammatory processes of the sinus.

- However, in the absence of bony destruction, these are nonspecific findings for diagnosing clinically significant sinusitis requiring antibiotic therapy

Treatment

- severe symptoms: antibiotics are given in concert with intranasal corticosteroids, as well as nasal decongestants.

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- DOC: Amoxicillin
  Resistant cases: then amoxicillin-clavulanate (Augmentin) is used.

- In mild cases, antibiotics are not normally used unless then symptoms have lasted for longer than 10 to 14 days.

PHARYNGITIS

00:12:27

- Pharyngitis is inflammation of the throat caused primarily by viruses.
- Approximately 10% of cases of pharyngitis are caused by S. pyogenes

- Streptococcal pharyngitis (strep throat) is important because poststreptococcal immune sequelae, such as rheumatic fever, may occur.

Clinical Manifestations

- Sore throat worsens on swallowing.

- Fever may also be present.

- Typical symptoms a/w an upper respiratory tract infection rhinorrhea, sinus tenderness, ear pain, cough may accompany the sore throat

- On examination:
  - An inflamed pharynx
  - Tonsils
  - Palate
  - A grayish exudate is often present on the tonsils
  - Tender, anterior cervical lymphadenopathy
  - Petechiae on the palate may also be a diagnostic clue for GAS

- S. pyogenes → most important bacterial cause.
  Group C and G streptococci also associated

- Pharyngitis caused by Neisseria gonorrhoea is likely to be the result of sexual activity and if it occurs in children, is

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considered as a sign of child abuse.

- Mycoplasma pneumoniae, Chlamydia pneumoniae and Arcanobacterium haemolyticum also cause pharyngitis.
- In certain countries where the diphtheriae vaccine is not widely used, Corynebacterium diphtheriae is a significant cause of pharyngitis, often accompanied by a pseudomembrane.
- Fusobacterium necrophorum, a gram -ve anaerobe, can cause pharyngitis accompanied by septic thrombophlebitis (Lemierre's syndrome)
- Note: although S. pneumoniae and H. influenza colonize the oropharynx, they do not cause pharyngitis.

Viruses

- Most cases of pharyngitis are caused by respiratory viruses, such as Adenovirus, Influenza A & B viruses, Parainfluenza viruse, Rhinovirus and Coronavirus.

- Other viral causes include: Coxsackie virus (herpangioma), Epstein-Barr virus (infectious mononucleosis), and herpes simplex virus, especially type 1.

- HIV causes and acute retroviral syndrome that includes pharyngitis as one of its components

Diagnosis

- Throat swab
- Tonsillar exudates
- Tender anterior cervical Lymphadenopathy
- Fever and absence of cough.
- Rapid antigen detection tests for GAS
- A throat culture is the most reliable method of determining whether S. pyogenes is the cause

Treatment

- Penicillin G, penicillin V or amoxicillin
- In penicillin allergic patients → erythromycin or cephalexin can be used.

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Prevention

- Vaccine against C. diphtheriae and Influenza virus
- Long-term carriers of GAS should not be treated
- Note that children who have RHD should receive penicillin orally for many years to prevent infection by S. pyogenes. Which could cause a flare of their RHD

COMMON COLD

00:21:11

- Viral infection of the upper respiratory tract, including some or all of the following structures: the Nose, Throat, Sinuses, Eustachian Tubes, Trachea and larynx.

Pathophysiology

- The viruses that cause the common cold are transmitted primarily by aerosols generated by sneezing or by direct contact.
- Direct contact involves either hand-to hand contact or hand-to-surface contact (Rhinovirus & Adenovirus).

Clinical manifestations

- Nasal congestion
- Decreased sense of smell
- Rhinorrhea (watery nasal discharge without purulence).
- Sneezing
- Malaise and sore throat.
- Headache

Pathogens

- Rhinovirus (> 100 serotypes) are the most common etiology (up to 50%).
- Coronaviruses
- Adenoviruses
- Enteroviruses → Coxsackie viruses

Diagnosis

- The common cold is usually diagnosed clinically.
- Erythematous and edematous nasal mucosa is seen on physical examination.
- Conjunctival and pharyngeal infection

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Treatment →Symptomatic

- Zing acetate in doses › 75 mg/day may reduce the duration of symptoms
- Other strategies: oral decongestants and buffered hypertonic saline nasal irrigation.
- If used for more than a few - days, nasal sprays can be used.

CROUP

00:25:35

- Croup is an inflammation of the larynx, trachea, and large bronchi (laryngotracheobronchitis).

Clinical Manifestations

- Inspiratory stridor is the key finding, together with a barking cough and a hoarse voice
- Symptoms may begin in a subtle fashion with nasal irritation and congestion and then rapidly progress to stridor over a day.

Pathogens

- Parainfluenza viruses, especially type 1 are the most common cause.
- Respiratory syncytial virus } 1% to 10% of cases
- Influenza virus } 1% to 10% of cases

Diagnosis

- The diagnosis is usually made clinically. Plain radiographs may show a 'steeple sign"
- Subglottic tracheal narrowing results in an inverted "V" shape

Treatment

- Patients with moderate to severe symptoms may be given corticosteroids →dexamethasone, with or without epinephrine
- There is no antiviral drug therapy

LARYNGITIS

00:27:26

- Laryngitis is inflammation of the vocal folds of the larynx.

Clinical manifestations

- Hoarseness
- Inability to speak (aphonia)

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- Laryngitis may be accompanied or preceded by an upper respiratory infection.

Etiology

- MCC → Parainfluenza viruses and Rhinoviruses
- Other causes:
  - Influenza virus
  - Adenovirus
  - Coronavirus
  - S. pyogenes
  - M. catarrhal's.
  - H. influenzae

Diagnosis → Clinically

Treatment → Hydration and voice test

→ Antibiotics are not needed

EPIGLOTTITIS

00:28:39

- Epiglottitis is an inflammation of the epiglottis.

Clinical Manifestations

- Rapidly worsening sore throat
- Odynophagia (pain on swallowing)
- Dysphasia (difficulty in swallowing)
- Pain may be out of proportion to physical examination findings.
- Airway obstruction can occur in severe cases

Treatment

- Epiglottitis in young children should be treated as a medical emergency

Pathogens

- MCC H. influenzae type B
- Less common pathogens include other H. influenzae, S. pneumoniae, S. pyogenes and S. aureus

Diagnosis

- Is made by visualization of the epiglottis.
- If indirect laryngoscopy (done primarily in children) is

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performed, a swollen and erythematous "cherry-red" epiglottis may be visualized.

- On lateral plain X-rays an enlarged epiglottis may be seen as a "thumb" sign

Microorganisms causing URTI and associated symptoms 00:31:00

Rhinitis common cold	Pharyngitis (sore throat), and tonsillitis	Laryngitis	Laryngotracheobronchitis
MC caused by viruses: • Rhinovirus • Coronaviurs • Adenovirus • Influenza virus • Human metapneumovirus • Respiratory syncytial virus	Symptoms: Pharynx and/or tonsils become inflamed, red, swollen, and show exudates, and sometimes a membrane is formed Viruses: (MC causes) • Influenza virus • Parainfluenza virus • Coxsackievirus A • Rhinovirus • Coronavirus • Epstein-Barr virus • Adenoviruses	Symptoms: • Hoarseness of voice • Lowering & deepening of voice Mostly viral agents: • Influenzae virus • Parainfluenzae virus • Rhino virus • Adeno virus • Corona virus • Human metapneumo virus	Age- Children, < 3 years age Symptoms: • Inspiratory stridor (high-pitched breath sounds resulting from turbulent air flow in the larynx) • Hoarseness • Fever • Cough (harsh, barking non-productive) Agents: • Parainfluenza virus (MC) • Influenza virus • Respiratory syncytial virus • adenoviruses
Sinusitis; Inflammation of the sinuses (d/t allergy or infections) Symptoms: • headache/ facial pain • thick nasal mucus	Bacteria: • streptococcus pyogenes (MC bacterial cause) • streptococcus group C & G • mycoplasma pneumonia • Vincent angina: Borrelia vincentii Leptotrichia buccalis Fungal: Candida albicans	If membrane or exudates present: • Streptococcus pyogenes • C. diphtheriae • Epstein-Barr virus	Epiglottitis Edema and inflammation of epiglottis & soft tissue above vocal cords Age: Children 2-6 years Symptoms: • Fever • Difficulty in swallowing • Inspiratory stridor MC agent: Haemophilus influenza type b

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Chapter 29

Lower Respiratory Tract Infections

Introduction

00:00:19

- Lower respiratory tract infections are an important cause of morbidity and mortality worldwide in children and in adults.
- Anatomic approach to lower respiratory tract infections, moving from the large bronchi (bronchitis) down to the very small bronchioles (bronchiolitis) and then into the alveoli where pneumonia occurs

Bronchitis

00:01:09

- It is self-limited inflammation of the bronchi
- Acute bronchitis must be distinguished from chronic bronchitis in which patients have a cough for more than 3 months.
- The coughing so characteristic of bronchitis is an attempt to clear the mucus produced by the inflammatory response to viral infection.
- Bronchitis occurs more often in the winter months than in the summer.
- Smoking predisposes to bronchitis (and pneumonia) by damaging the cilia in the bronchia, leading to an inability to clear mucus from the respiratory tract

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- Cough is the most prominent symptom of bronchitis.initially, bronchitis presents with the symptoms of an upper respiratory infection, namely, nasal congestion, scratchy sore throat and perhaps a low-grade fever.

- Physical examination typically reveals expiratory wheezes However, if cough persists for > 5 days & pneumonia has been ruled out, acute bronchitis should be suspected

- Bronchitis is self-limited & usually resolves in 1 to 2 wks. However, cough may persist for several more wks d/t airway hyperreactivity

- Respiratory viruses are the MC pathogens (Influenza A &, parainfluenza virus, coronavirus, rhiovirus, respiratory syncytial virus [RSV], and human metapneumovirus).

- Bacterial pathogens are not thought to play a significant role in acute bronchitis

- The diagnosis is primarily made clinically.

- Cough, with or without sputum production, which may persist for > 5 days, is the typical presentation.

- Patients are usually afebrile but may have a low-grade fever. Suptum are typically not done.

- In patients with chronic cardiorespiratory disease, a rapid antigen test for influenza virus may be useful because oseltamivir can shorten the duration & intensity of symptoms

- Because treatment of both upper respiratory infections & acute bronchitis (usually viral) from pneumonia (mainly bacterial), which does require antimicrobial therapy.

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- A chest radiograph may be performed to determine whether pneumonia is present
- Treatment involves reassurance and symptom relief with agents such as nonsteroidal anti-inflammatory drugs
- The neuraminidase inhibitor oseltamivir (Tamiflu) should be given to unimmunized individuals with chronic cardiorespiratory disease
- Handwashing is recommended to reduce the carriage of respiratory viruses

Bronchiolitis

00:08:30

- Bronchiolitis is inflammation of the bronchioles - the small airways < 2 mm in diameter
- Bronchiolitis among infants & young children is primarily infectious
- Particularly among children under 2 yrs of age viruses can directly damage the epithelial cells of the terminal bronchioles, causing inflammation & obstruction of the small airways. Prematurity is an important predisposing factor
- Usually children initially have symptoms consistent with an upper respiratory tract infection and then are noticed to have increased respiratory distress
- Children under 2 yrs old in particular may have tachypnea, wheezing, nasal flaring and chest restractions.
- In severe cases, hypoxia, apnea and respiratory failure may ensure. In most cases, recovery occurs in 1 to 2 wks
- RSV is the MC pathogen. Other etiologies include influenza virus, parainfluenza virus, adenovirus, coronavirus, rhinovirus and

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idiopathic causes

- Bronchiolitis caused by RSV occurs primarily in the winter months

- The diagnosis is primarily clinical Upper respiratory tract infection symptoms followed by lower respiratory tract symptoms and signs (e.g., nasal flaring, wheezing) in a young child during the winter would be very suggestive of bronchiolitis.

- Chest radiograph typically shows hyperinflation of the lungs.

- An enzyme immunoassay (EIA) for RSV antigen in respiratory secretions is available for diagnosis in hospitalized patients.

- A PCR assay that detects the RNA of RSV is also available.

Rx

- Self-limited disease in most cases, general supportive measures are adequate in most cases

- Patients with moderate or severe respiratory distress will require hospitalization.

- Ribavirin delivered by aerosol into the lungs is approved for severe disease caused by RSV, but it's use is limited to hospitalized infants. Inhaled bronchodilators may be useful

- Hand washing to minimize transmission of pathogens is an important strategy

- Pallivizumab is a monoclonal antibody against the RSV F (fusion) envelope protein that may be used in certain populations to decrease the risk of disease caused by RSV

PNEUMONIA

00:16:58

- Pneumonia is an inflammation of the lung affecting the alveoli.

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- Pneumonia is community acquired Vs hospital acquired to help us determine the spectrum of potential pathogens that differs based on setting

- Empiric therapy is often given in pneumonia because of this reason

- Hospital-acquired pneumonia, also known as nosocomial pneumonia, is pneumonia that occurs 48 hours or more after admission to the hospital and was not present at the time of admission.

- The alveoli of the lungs are continually exposed to microbes from the environment via the upper respiratory tract.

- Our host defenses usually keep these potential pathogens in check. However, disease can occur when there is a particularly virulent organism, when there is large burden of organisms inhaled from the environment or aspirated from the oropharynx, or when there is a defect in host immunity.

Predisposing factors to pneumonia:

- Extremes of age
- Chronic obstructive pulmonary disease (COPD)
- Chronic bronchitis
- Diabetes mellitus
- Cystic fibrosis
- Congestive heart failure."
- IV drugs users
- Alcoholics
- Seizure

} have a high risk of pneumonia because they can aspirate organisms into the lung when unconscious

- People exposed to water aerosols, especially from air conditioners, are at risk for pneumonia caused by Legionella

- Hospitalized patients in the ICU are at risk for ventilator-

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associated pneumonia by Gram -ve rods eg E. coli, Pseudomonas & Acinetobacter

Clinical Manifestations

Symptoms:

- Cough with sputum
- Fever with chills
- Chest pain
- Shortness of Breath
- Rusty sputum
- Currant Jelly sputum

Physical examination

- Tachypnea
- Rales
- Rhonchi
- Dullness on percussion → Lung is consolidated

- Patients who are intubated and who acquire a nosocomial pneumonia may only have fever as a presenting sign

- Pneumonia may be complicated by an infected pleural effusion or a pleural empyema

- A pleural empyema is walled-off collection of pus in the pleural space

Pathogens

- MCC of community-acquired pneumonia Streptococcus pneumoniae
- Other common bacterial pathogens:
  - Klebsiella, Pneumoniae and H. influenzae.

Note: Nontypeable strains of H. infuenzae that cause pneumonia in

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elderly patients with COPD rather than type B strain

- Other pathogens:
  - Mycoplasma pneumoniae, Legionella species, and Chlamydophila pneumoniae
- Infection with MTB can also manifest as pneumonia.

Note: Approximately 30% of adults with community acquired pneumonia, no pathogen, neither bacteria nor virus is isolated

- Causes of pneumonia in a neonate are those acquired during passage through the birth canal.

- The main cause of pneumonia in an infant, Chlamydia trachomatis is also acquired during passage through the birth canal but is a less aggressive pathogen so its onset in delayed.

Note: M. pneumoniae is the MCC in young adults

Age	Bacteria	Viruses
Neonates	GroupB streptococci E. coli	Respiratory syncytial virus (RSV)
Infants	C. trachomatis S. pneumonia	RSV Parainfluenza virus
Children	S. pneumoniae Haemophilua influenzae	RSV Parainfluenza virus
Young adults	M. pneumoniae C.pneumoniae S. pneumoniae	Various respiratory Viruses (eg. adenovirus)
Older adults	S. pneumonia H. influenzae Legionella pneumophile	Influenza virus

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Predisposing factors	Typical Pathogens
Alcoholism	Klebsiella pneumonia
Bird exposure (psittacosis)	Chlamydophila psittaci
COPD including smoking related	H. influenzae
Cystic fibrosis	Pseudomonas aeruginosa
Imported wool, spores in wool (wool-sorter's disease)	B. anthrax
Influenzae virus infection	S. aureus
Intubation, postsurgery and ICU	Coliforms: P. aeruginosa, S. aureus
Mouse dropping exposure, especially in southwestern states	Hantavirus
Sheep exposure, especially tissue (Q fever)	C. burnetii
Travel to or reside in central valley of California, Arizona, or New Mexico	Coccidioides immitis
Travel to or reside in Ohio or Mississippi river valleys	Histoplasma capsulatum
Ventilator-associated, especially in ICU	Acinetobacter species

Diagnosis

- The "gold standard" for a diagnosis of pneumonia is an infiltrate on a plain chest radiograph
- Clinical data may help, but ultimately the chest radiograph is the most important diagnostic tool
- Sputum analysis for Gram stain and culture and blood cultures may be helpful in the hospitalized patient but are only optional in an outpatient setting because therapy is largely empiric for community acquired pneumonia
- In pneumonia caused by one of the encapsulated pyogenic

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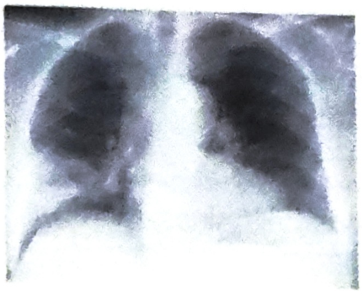
bacteria such as S. pneumoniae

- White blood cell count is frequently elevated and the number of neutrophils is often increased



Lobar pneumonia caused by S. pneumoniae

Area of consolidation in right lung

CURB-65 SCORE

00:42:33

The prediction of prognosis of CAP in an adult is done by CURB-65 score

- C (Confustion) = 1 point
- U (blood urea nitrogen > 19 mg/dL) = 1 point
- R (respiratory rate > 30 mint) = 1 point
- B (BP < 90/60) = 1 point
- 65 (Age > 65 yrs) = 1 point
- Higher the score, greater is the mortality
- If the score < 1, outpatient therapy is indicated
- If the score > 1, patient should be hospitalized

Clinical Pulmonary Infection Score (CPIS) used for ventilator associated pneumonia (VAP)

00:45:35

Parameter (s)	Score 0	Score 1	Score 2
Temperature (°C)	≥36.5°C & ≤38.4°C	≥38.5°C & ≤38.9°C	≥39°C & ≤36.4°C
Leukocytosis	≥4000 & ≤11,000	< 4000 & > 12,000	
Tracheal aspirate	None	Non-purulent	Purulent
Oxygenation (PaO_2/FIO_2 mmHg)	> 250 or ARDS		≤ 250 & no ARDS
Chest radiograph	No opacity	Diffuse (patchy) opacity	Localized opacity
Progressive radiological progression	No radiological progression		Radiological progression
Culture of tracheal aspirate	Pathogenic bacteria light or no growth	Pathogenic bacteria moderate of heavy growth	
Culture - Gram stain	Different morphology	Same morphology as Gram	

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- It is important that sputum (not saliva) be sent to the lab for Gram stain and culture.
- If the specimen contains many neutrophils and few epithelial cells, then the specimen is likely to be sputum and will be analyzed
- If, however, the specimen contains many epithelial cells and few neutrophils, then the specimen is saliva and will be rejected by the lab

Identification features/ detection methods of common organisms causing pneumonia

Agents of pneumonia	Direct demonstration in sputum	Culture identification
Lobar pneumonia		
Streptococcus pneumonia	Pus cells > 25/ LPF and epithelial cell < 5/ LPF Gram +ve cocci in pair, lanceolate shaped	• Alpha haemolytic, draughts man-shaped colonies on blood agar, sensitive to optochin • Bile soluble, ferments insulin
Haemophilus influenzae	Pleomorphic Gram −ve bacilli	Satellitism on blood agar with S. aureus streak line
S. aureus	Gram +ve cocci in clusters	BA: golden yellow haemolytic colonies catalase +ve Coagulase +ve
Gram −ve bacilli: E. coli, Klebsiella, Pseudomonas	Gram −ve bacilli	Identification is based on: • Growth on MacConkey agar (LF or NLF colonies) • Biochemical reactions (ICUT: Indole, Citrate, Urease, TSI)

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PrepLadder

Interstitial or atypical pneumonia		
Chlamydophila pneumoniae	Direct immunofluorescence test Antigen detection by enzyme immunoassay Nucleic and amplification test (NAAT) detecting specific genes	Serology-antibody detection by • CFT using LPS antigen • ELISA using recombinant LPS antigen • Micro- IF test using outer membrane protein antigen
Mycoplasma pneumoniae	Direct immunofluorescence test Capture ELISA – detecting antigen (P1 adhesin) PCR targeting P1 adhesin gene	Cuture- fried egg colonies on PPLO agar Antibody detection • Non-specific test (cold agglutination test) • Specific test (e.g. ELISA)
Legionella pneumophila	Pus cells › 25/LPF & epithelial cells ‹ 5/LPF Detection of specific antigen in sputum, urine	Growth on BCYE medium
Viral pneumonia	Detection of specific viral antigen in sputum Detection of specific viral genes in sputum (PCR)	

- Treatment for community-acquired pneumonia is largely empiric because microbiologic diagnostic strategies are generally insensitive.

- OPD Rx:

Macrolides →Azithromycin

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Tetracycline → Doxycycline

Respiratory Quinolone → Levofloxacin

- Inpatients: Ceftriaxone + Macrolide/Respiratory Quinolone Monothrerapy

- Patients with suspected hospital-acquired pneumonia may be given broader spectrum agents such as a Carbapenem depending on the local epidemiology, given that many hospital-acquired infections are multidrug resistant .

- Prompt initiation of antibiotics is important because morbidity and mortality increase after a delay of more than 8 hours

- Drainage of an empyema or infected pleural fluid should be performed

- The influenza vaccine is effective in decreasing the likelihood of pneumonia.

- The Pneumococcal Polysaccharide (nonconjugate) Vaccine available for older adults is important in decreasing bacteremia associated with pneumonia and mortality but not the likelihood of pneumonia itself.

- Because it is the Nontypeable strains of H. influenzae, rather than the type B strain, that are the MCC of pneumonia, the conjugate vaccine against type B is not an important source of protection.

- Smoking cessation and treatment of alcohol abuse may also decrease pneumonia risk

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PrepLadder

Organism Causing Lower Respiratory Tract Infections (LRTI) 01:05:40

Pneumonia		Bronchitis
Community acquired		Hospital acquired
No co-morbidity: • Streptococcus pneumoniae (MC) • Atypical pathogens: – Chlamydophila pneumoniae and C. psittaci – Legionella and Mycoplasma – Coxiella burnetti (Q fever) – Viral pneumonia (influenza, adenovirus, parainfluenza, RSV) Co-morbidity: • Alcoholism: S. pneumoniae, H. influenzae • COPD: H. influenzae, M. catarrhalis, S. pneumoniae • Post – CVA-aspiration: S. pneumonia	Bacteria agents: • Gram –ve bacilli (MC) – MDR non-fermenters (Pseudomonas & Acinetobacter) – MDR Enterobacteriaceae E. coli, Klebsiella & Enterobacter • Staphylococcus aureus (both MRSA & MSSA) • S. pneumoniae (rarely, in early stage) Viral agents: • Influenza, adenovirus, parainfluenza RSV	Bacterial agents: • B. pertussis • B. parapertusis • M. pneumoniae • Chlamydophila Viral agents: • Influenza virus • Adeno viruses • Rhino viruses • Corona viruses Bronchiolitis: Viral agents: • Respiratory syncytial viruses • Parainfluenza virus • Rhino virus • Influenza virus • Adeno virus • Entero virus • Human metapneumo virus

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PrepLadder

- Post-obstruction of bronchi: S. pneumoniae, anaerobes
- Post-influenza: S. pneumoniae, S. aureus
- MCC of CAP → S. pneumoniae
- MCC of Atypical Pneumonia → M pneumoniae

LUNG ABSCESS

01:15:02

- Lung abscess in a necrotic process within the lung parenchyma that frequently results in a cavity with an air-fluid level
- Patients may aspirate oropharyngeal bacteria into the lower airways and alveoli. This usually occurs when the patient is in the recumbant position and cannot clear secretions.
- For example, aspiration can occur when a person is unconscious from drug overdose, excess alcohol intake, or the anesthesia that accompanies surgery
- Poor oral hygiene is a common predisposing factor. A pneumonitis may first occur, but this can progress to necrosis in a week or so.
- A lung abscess caused by S. aureus may infect the lung via the bloodstream from a distant site of infection such as right-sided endocarditis in an intravenous drug user
- Patients present with symptoms typical of pneumonia with fever and productive cough.
- The sputum is often foul smelling, indicating the presence of anaerobes
- These symptoms may be indolent and progress over a period of wks. Systemic symptoms such as night sweats, fatigue and weight loss may also be present

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PrepLadder

- The MC organisms are anaerobes or mixed aerobes and anaerobes that are part of the oral flora.

- Anaeorbes commonly involved → Peptostreptococcus species, Prevotella species, and Fusobacterium nucleatum.

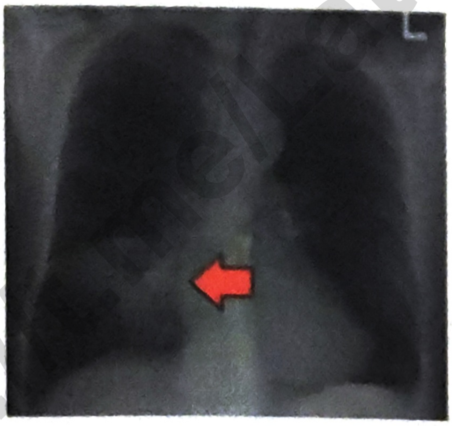
- Aerobes include Streptococcus milleri and S. aureus. (Clinicians often use the term "aerobe" rather than facultative to describe bacteria that are not anaerobic.)

Diagnosis

- A chest radiograph shows a pulmonary infiltrate with a cavity, often with an air-fluid level

- An air-fluid level occurs when the abscess erodes a bronchus and some of the pus in the abscess is coughed up and replaced by air

- Pleural fluid, if present, and blood culture may provide microbiologic data, but anaerobes may be difficult to identify



Lung Abscess

Arrow points to air-fluid interface within the abscess.

Treatment

- Clindamycin or ampicillin-sulbactam options → 4-6 wks
- Patients who do not respond to antibiotics will required surgical drainage

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1. A 8 yr old girl admitted with complaints of productive cough chest pain, shortness of breath since 3 days

O/E:

On Percussion: Dullness over left sixth ISC

On Auscultation: Crepitation & Rales over sixth ICS

On Chest X ray: Homogeneous Ground Glass Opacities in the left lower lobe

Sputum is collected & subjected to microscopy & Culture

What is the Diagnosis?

→ Typical Case of PNEUMONIA

Productive cough
Ground glass opacities } LRTI
Crepts & rales

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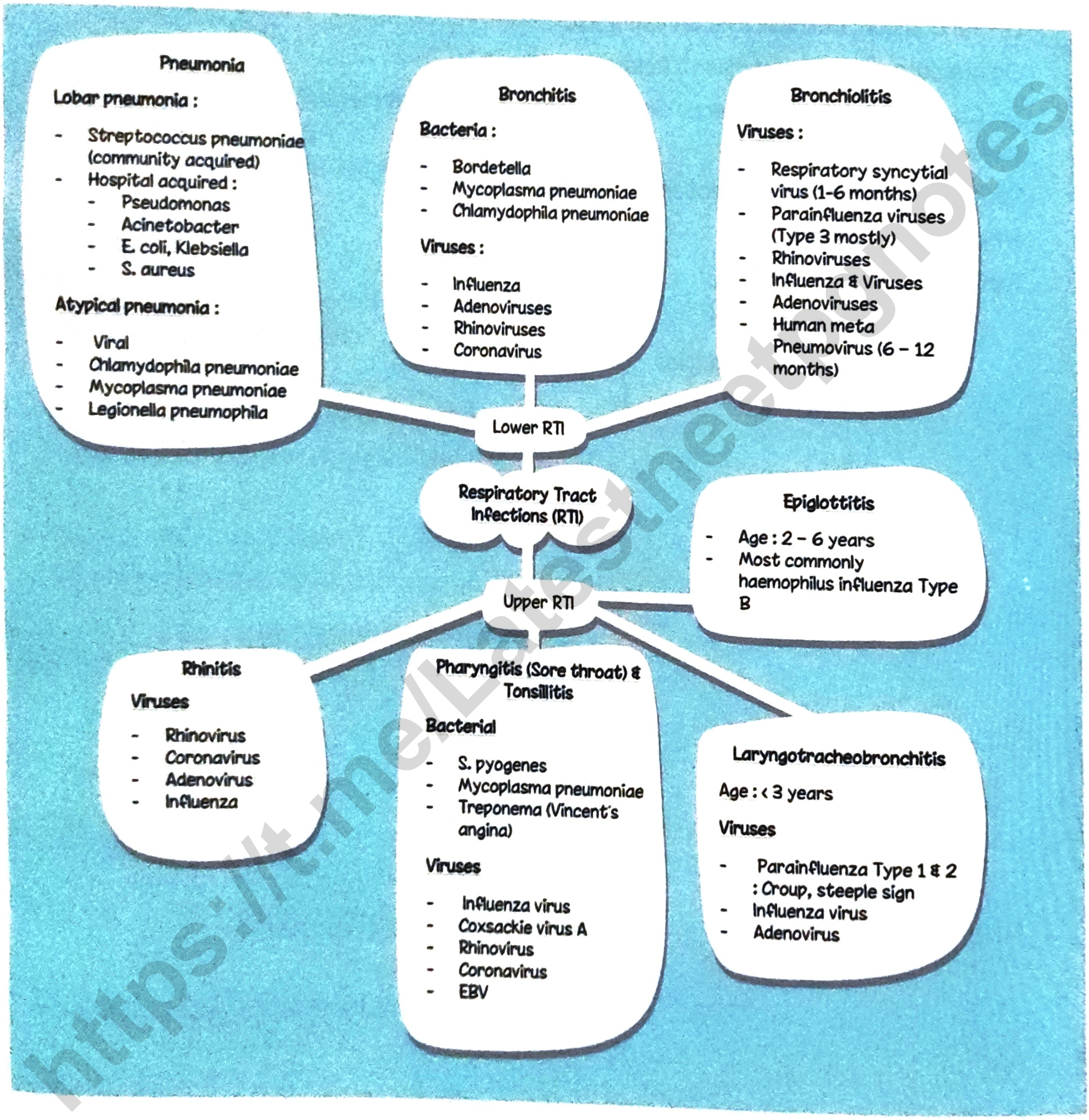
PrepLadder



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Chapter 30

CENTRAL NERVOUS SYSTEM INFECTIONS

Introduction

00:00:13

- CNS infections are often life-threatening and can have severe sequelae
- Leads to inflammation and edema → damage to brain tissue & loss of function
- MCC: Bacteria, Virus
- Others: Fungi
  Protozoa
  Helminthes

MENINGITIS

00:02:00

Meningitis is an infection of the meninges, the membranes that line the brain and spinal cord

Types (Depending on speed of the initial presentation and the rate of progression of the illness)

- Acute
- Subacute
- Chronic

Acute Meningitis

00:03:05

Etiology:

- Bacteria →S. pneumoniae and N. meningitides
- Virus →Coxsackie virus and HSV type 2

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PrepLadder

Viral Meningitis

- Also know as aseptic meningitis

Subacute meningitis

Etiology

- MTB
- Fungi → Cryptococcus

Pathophysiology

- Hematogenous spread (MC)
- Direct spread via adjacent infections
- Via neurosurgery → CSF shunt
- Via trauma

Acute bacterial Meningitis

Nasopharyngeal

↓ f/b

Hematogenous spread

↓

Edema

↓

↑ CSF

↓

- Headache
- Cerebral vasculitis
- Cerebral infarction

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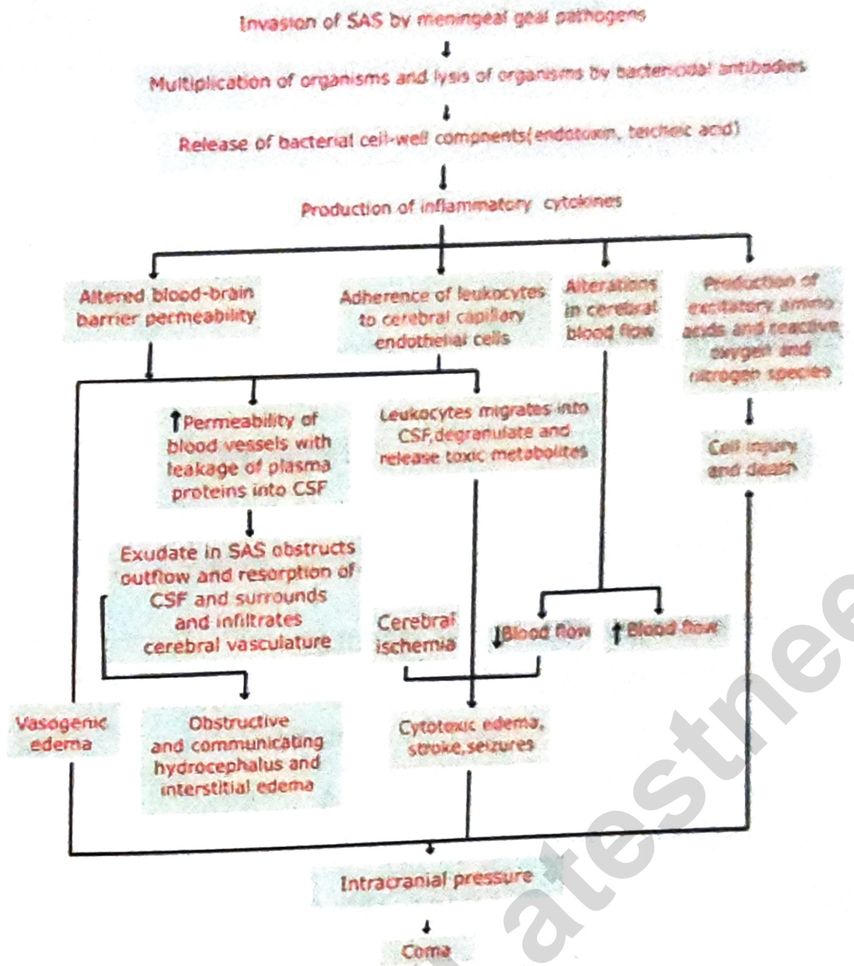
Pathogenesis of Bacterial Meningitis



Clinical Manifestations

Early symptoms → Fever, headache, stiff neck & photophobia

↓ If Untreated

Vomiting, seizures, focal neurologic deficits & altered mental status

N. meningitidis infection can be associated with Rash → meningococcemia a/w purpura fulminans

Purulent Meningitis

Purpura Fulminans

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PrepLadder

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Or click this page to join

- MCC of pyogenic meningitis → S. pneumoniae
  - Meningococci (25%)
  - S. agalactiae (15%)
  - Listeria (10%)
  - H. influenzae (<10%)

- Neonates: S. agalactiae
  GNB → E.coli, Klebsiella and listeria

- Elderly (>60 yrs): S. agalactiae
  L. monocytogenes

- Less common pathogens: Borrelia burgdorferi (Lyme disease)
  Treponema pallidum (syphilis)

Acute Viral pathogens

00:21:14

- MCC → Enterovirus (Coxasackie virus & Echovirus)
  - Occurs primarily in young children
  - Peak in summer season

Note: HSV-2 causes meningitis
HSV-1 causes encephalitis

- Primary genital infections with HSV-2 are more likely to result in meningitis than Recurrent HSV-2 infections

Other Viruses a/w Meningitis:

- Varicella zoster virus
- Arboviruses → West Nile Virus & St. Louis encephalitis virus
- Mumps virus

Suacute and Chronic Meningitis

00:25:05

- MCC: MTB
  Fungi → Cryptococcus, Coccidioides & Histoplasma

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PrepLadder

Normal CSF Findings

- CSF pressure : 50-150 mm of water
- Total leukocyte count : 0-5/mm^3
- Predominant Cell Type : Lymphocytes
- Glucose : 40-70mg%
- Total proteins : 15-45 mg%

CSF analysis in pyogenic meningitis

Opening pressure	>180 mmH_2O
White blood cells	10/μL to 10,000/μL; neutrophils predominate
Red blood cells	Absent in nontraumatic tap
Glucose	<2.2 mmol/L (<40 mg/dL)
CSF/ serum glucose	<0.4
Protein	>0.45 g/L (>45 mg/dL)
Gram's stain	Positive in > 60%
Culture	Positive in > 80%
PCR	Detects bacterial DNA
Latex agglutination	+ve in S. pneumoniae, N. meningitides, H. influenzae type b, E. coli, group B streptococci
Limulus lysate	+ve in cases of gram –ve meningitis

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Identification features/ detection methods of common organisms causing meningitis

Agents of meningitis	Biochemical analysis	Direct demonstration	Culture identification
S. pneumoniae	Pyogenic meningitis • CSF pressure: highly elevated >180 mm of water • TLC: highly elevated, neutrophilic • 100-10,000/mm^3 • Glucose: ↓ed to absent <40mg/dL • Total proteins: >45mg/dL	Gram +ve cocci in pair, lanceolate shaped	• Alpha hemolytic draughtsman-shaped colony on BA • Sensitive to optochin • Bile soluble • Ferments insulin
S. agalactiae		• Beta hemolytic pin point colony on BA • CAMP test +ve, resistant to bacitracin	
Neisseria meningitis		Oxidase +ve, Catalase +ve growth on chocolate agar	
H. influenzae		Pleomorphic gram -ve bacilli	Satellitism on BA with S. aureus streak line, growth surrounding disk containing combined X and V factors
E. coli or other Gram -ve bacilli		Identification is based on colony grown on MacConkey agar and biochemical reactions	

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PrepLadder

Cryptococcus neoformans True yeast	• CSF pressure: slightly elevated/ normal • Leukocyte count: slightly elevated & lymphocytic • Glucose: normal • Total proteins: normal or slightly elevated	• India ink stain shows budding yeast cells • Latex agglutination test detects capsular Ag in CSF	BA or CA or SDA shows mucoid white colonies
Viral meningitis		Detection of DNA or RNA in CSF by PCR or RT PCR	
Tubercular meningitis	• CSF pressure: moderately elevated, cobweb coagulum seen • Leukocyte count: moderately elevated & lymphocytic • Glucose: slightly ↓ed • Total proteins: moderate to markedly ↑ed	• ZN stain of CSF shows acid fast bacilli • Detection of specific genes in CSF by PCR or GeneXpert	• Growth on LJ medium → rough, buff, tough colonies • Growth on MGIT

* Lumbar puncture should not be performed if there are increased signs of ICT eg. Papilledema or focal neurologic signs, as can lead to herniation of brainstem & death may occur

00:49:12

Treatment

- Empiric therapy for acute bacterial meningitis
- Drugs with excellent penetration to the CSF & that are bactericidal are used

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PrepLadder

- Ceftriaxone/ cefotaxime + vancomycin (older children & adults)
  - Ampicillin shoud be added if listeria is a likely cause
- Empiric therapy for neonatal bacterial meningitis
  - Ampicillin + ceftriaxone or cefortaxime
- Acyclovir is used for the treatment of HSV and VZV infection

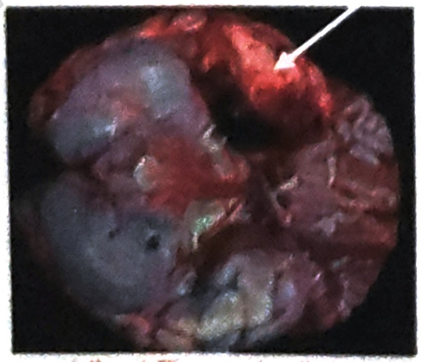
Prevention

- Immunization
- Pre exposure and post exposure chemoprophylaxis
- Vaccines: →Conjugate vaccines
  - → Pneumococcal vaccine (Protects against 13 serotypes)
  - →Meningococcal vaccine (Protects against 4 serotypes)
  - →H. influenzae vaccine (Protects against only type B serotype)
- Pre exposure chemoprophylaxis against S. agalactiae to reduce vaginal carriage in mother
- Post exposure chemoprophylaxis aimed at reducing nasopharyngeal carriage of N. meningitidis and H. influenzae type B.
- Close contacts of patients with meningitis should receive ciprofloxacin for Neisseria or Rifampicin for Haemophilus

ENCEPHALITIS

00:54:07

- Encephalitis is an infection of the brain parenchyma predominantly caused by viruses
- Meningoencephalitis →when both brain and meninges are involved



Viral Encephalitis

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PrepLadder

Clinical Manifestations

- Fever
- Headache
- Altered mental status
- Seizures
- Focal neurological deficits
- Rabies encephalitis present with:
- Furious rabies (80%)
  - Hyperactivity
  - Agitation
  - Delirium
  - Hydrophobia
  - Seizures
- Dumb rabies (20%)
  - Paralytic symptoms
    - Ascending paralysis without hyperactivity
- Coma and death are common in both forms

Pathogens

- Viruses
  - HSV-1 (15%) } MCC
  - HSV-2 } Acyclovir
  - Arboviruses (5%)
  - VZV, CMV and EBV
- WNV MC arboviral cause of encephalitis is the US
- 80% WNV are asymptomatic.
- 20% WNV develop flu-like illness
- <1% develop CNS disease

Diagnosis

- Mild elevation of CSF lymphocytes
- Elevation of proteins
- Glucose normal
- PCR & ELISA is done

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PrepLadder

Dx of Rabies:

- Direct fluorescent Ab staining (biopsy of skin from nape of neck)
- PCR Assay (CSF, Saliva or Tissue)
  - Identifies animal reservoir & geographical location of virus

Treatment

- Rx of choice for HSV-1, HSV-2 & VZV → IV Acyclovir
- No Antiviral therapy for Arbovirus & Rabies Encephalitis
- To reduce the transmission of HSV-2 to the neonate, pregnant women with active lesions late in pregnancy should receive acyclovir and should be considered for CS

BRAIN ABSCESS

01:08:17

- A brain abscess is a localized, walled-off collection of pus surrounded by a fibrous capsule

Etiology

- MCC → bacteria
- Fungi and protozoa
- Viruses do not cause brain abscess

Pathophysiology

- Complication of head and neck pyogenic infections, such as sinusitis, otitis media, and dental infections
  - Sinusitis predisposes to lesions in the frontal lobe
  - Otitis media predisposes to lesions in the temporal lobe
- Hematogenous spread from an infected site (infective endocarditis) can also occur
- Increased use of immunosuppressive drugs, IV catheters, and hyperalimentation are a/w fungal brain abscesses
- Immunocompromised patients, especially those with AIDS, also have brain abscesses caused by Toxoplasma gondii

Clinical Manifestations

- ↑ICT → Headache, vomiting (in early course of disease)
- Focal neurologic deficits and seizures (as the lesion expands)

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PrepLadder

Pathogens

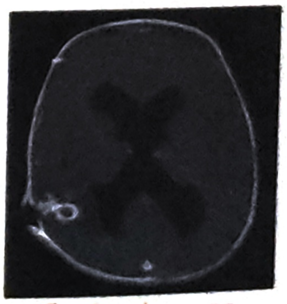
- Bacteria
  - Streptococci
  - Prevotella
  - Fusobacterium
  - Bacteroides

- Fungi
  - A. fumigatus
  - Mucor and Rhizopus species → Rhinocerebral mucormycosis
  - Cryptococcus → in patient's with HIV/AIDS
  - Candida

- Protozoa
  - T. gondii → a/w immunocompromised patients or with those receiving immunosuppressive drugs
  - MOT:
    - Solid organ transplant
    - Ingestion of raw meat containing cysts
    - Exposure to cat feces containing occytes
    - Transplacental transmission → intracranial calcifications in fetus

Diagnosis

- MRI → ring-enhancing lesion
- Culture of pus from abscess for bacteria and fungi
- Gram staining reveals several types of bacteria indicating a mixed infection



Brain abscess

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PrepLadder

MRI showing a small ring-enhancing lesion with mild surrounding edema adjacent to the ventricular catheter and ventricular dilatation

Treatment

- Empiric antimicrobial therapy for bacteria
  - Ceftriaxone or cefotaxime + metronidazole
- Treatment of bacterial and fungal brain abscess may require a surgical drainage of pus
- Treatment of Toxoplasma brain abscess includes a combination of pyrimethamine and sulfadiazine

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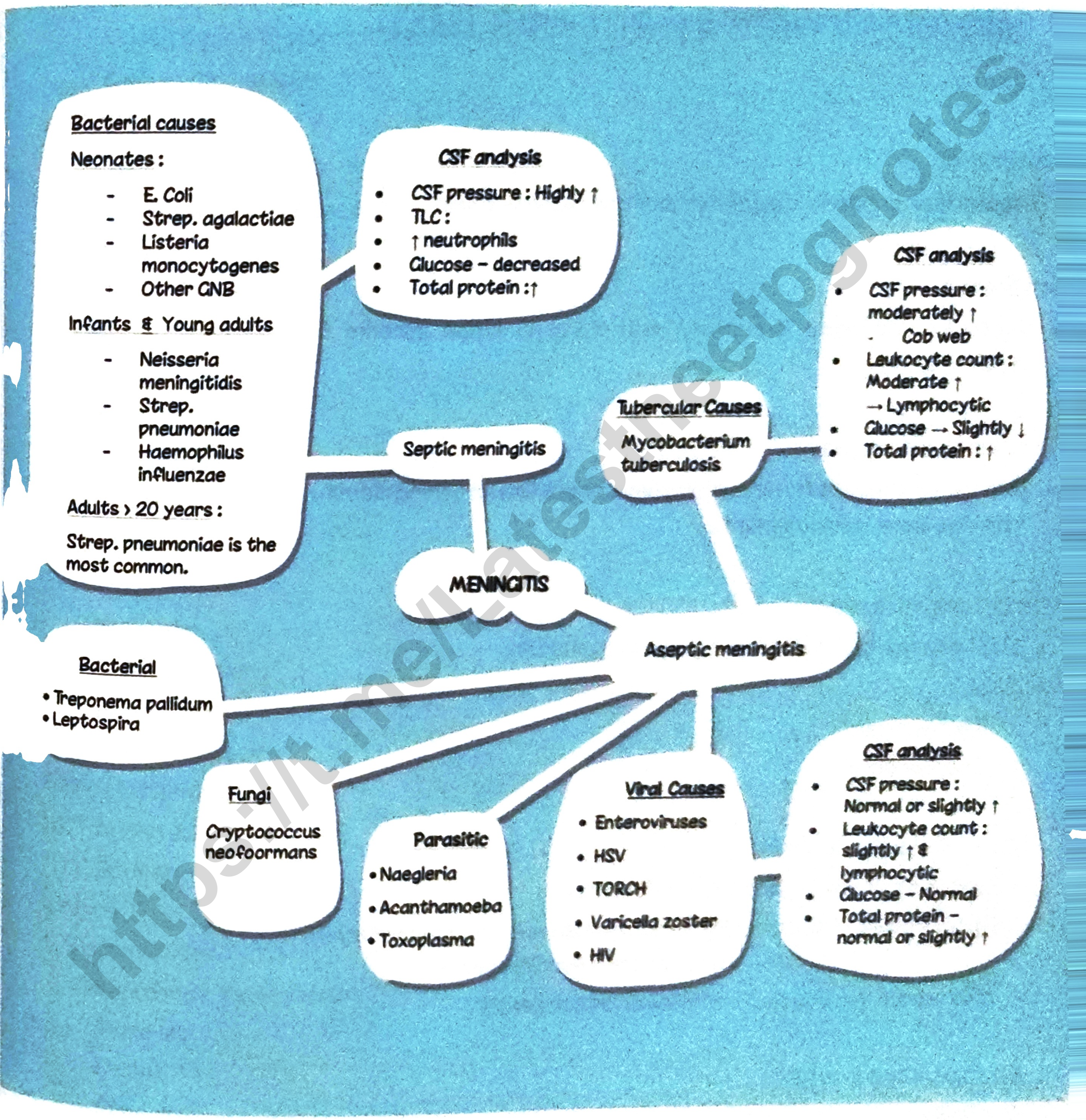

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PrepLadder

Chapter 31

Blood Stream Infections (BSI)

Bacteremia

- Presence of bacteria in blood without any multiplication

Septicemia

- Bacteria circulating & actively multiplying in blood stream & produce toxins

Transient Bacteremia

- Occurs spontaneously or after some event like brushing teeth, chewing food, instrumentation

Continuous Bacteremia

- Bacteria are in blood stream at a constant rate
- Seen in
    - Septic shock
    - Endocarditis

Intermediate Bacteremia

- Bacteria released in blood intermittently
- Eg. Undrained abscess → Bacteria are released approximately 45 mins before a febrile episode
- Also seen in Meningitis, Pneumonia, Osteomyelitis

Etiological agents of BSI

Bacteria

1. Salmonella typhi
2. Brucella

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PrepLadder

3. Leptospira, Borrelia (Spirochaetes)
4. HACEK (Endocarditis group)
5. Rickettsiae
   - Infect vascular endothelium
   - Doesn't grow in blood culture
   - Cultured in cell lines
6. GPC
   - Staphylococcus
   - β Hemolytic Streptococci
   - Enterococcus
   - Pneumococcus
7. GNC
   - Meningococci
8. GPB
   - B. anthracis
   - Listeria
9. GNB
   - E.coli
   - Klebsiella
   - Enterobacter
   - Pseudomonas
   - Acinetobacter
   - H.influenzae
10. Anaerobes
    - Bacterioides

Viruses

1. Retro viruses
2. Hemorrhagic fever viruses
   - Marburg
   - Ebola
   - Dengue
   - Chikungunya
   - Yellow fever
   - Herpes family → EBV, CMV

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PrepLadder

Parasites

1. Plasmodium
2. Babesia microti
3. T. gondii
4. Leishmania
5. Trypanosoma
6. Filarial parasites

Fungus

1. Candida albicans & Non Candida albicans (NCA)
2. Dimorphic fungus
   - Histoplasma capsulatum
   - Blastomyces
   - Coccidoides
   - Paracoccidiodes

Types of BSI

1. Intravascular
2. Extravascular

Pre disposing factors for BSI

- Immunosuppression
- Use of Broad spectrum antibiotics for a long time
- Invasive procedures (like Central line)
- Extensive surgeries

Intravascular BSI

- Infection of blood vessels
- Includes
  - Endocarditis
  - Myocarditis
  - Pericarditis

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PrepLadder

Extravascular BSI

- Organisms multiply at primary site such as lung (in case of Pneumonia)

↓

Drained by lymphatics

↓

Reaches blood stream

↓

Move to various organs

- Organisms ultimately reach Reticuloendothelial system (RES)

↓

Escape various mechanisms of RES & multiply

↓

Septicemia

Portal of entry

- Genito Urinary Tract (GUT) (25%) → MC
- Respiratory tract (20%)

Clinical manifestations of BSI

- BSI have Bacteremia stage f/b Septicaemia stage
- Clinical manifestations evident only in Septicemic stage

- Stages of BSI based on severity & extent of organ failure
  - Sepsis
  - Septic shock

Sepsis

- Defined as life threatening organ dysfunction caused by a dysregulated host response to infection
- Sepsis is diagnosed by SOFA (Sepsis related Organ Failure Assessment) Score

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PrepLadder

SOFA Score

- Have 6 parameters
  1. Respiratory system → PaO_2, FiO_2
  2. Coagulation system → Platelet count
  3. Liver → Serum bilirubin
  4. Cardivascular system → Mean Arterial Pressure (MAP)
  5. CNS → Glasgow Coma Scale (GCS)
  6. Renal system → Serum creatinine & Urine output

Organ dysfunction

- Identified as acute change in total SOFA score ≥2 points following the infection

qSOFA (Quick SOFA) Score

- Have 3 parameters
  1. Respiratory rate ≥22/min
  2. Altered mental status
  3. Systolic BP ≤100 mmHg

Septic Shock

- Defined as subset of Sepsis in which underlying circulatory & cellular / metabolic abnormalities are profound
- Patient with Sepsis shock are identified by clinical construct of Sepsis with
  - Persisting Hypotension requiring Vasopressors to maintain MAP (Mean Arterial Pressure) ≥65 mmHg
  - Serum Lactate level > 2 mmol/L (18mg/dL) despite adequate volume resuscitation
- Patients with Septic shock have a mortality of >40% in contrast to 10% for Sepsis cases

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PrepLadder

Lab Diagnosis

- Specimen Collection for Blood culture
  - Site
    - Blood for Culture → Always collected in pairs from 2 separate Venonpuncture & 2 separate skin decontamination processes
  - Preparation of the site
    - To avoid contamination with skin flora, blood should be collected under strict aseptic conditions using sterile disposable syringe
  - Skin Decontamination
  - Timing of collection
    - Blood collected before starting Antimicrobial therapy
  - Blood volume
    - Adult → 8-10 mL of blood per bottle
    - Pediatric → 1-3 mL of blood per bottle
    - Ratio of Blood : Blood culture fluid → 1:10
  - Number of blood cultures
    - Atleast 2-3 blood Culture sets are required
    - Each set consists of 2 bottles → 1 Aerobic & 1 Anaerobic
  - Dispensing
    - Collected blood is directly dispensed into Blood Culture bottle

Steps of Collection of Blood for Blood Culture

1. Perform hand hygiene & use sterile gloves
2. Apply tourniquet, palpate the vein & mark the area
3. Use 70% Alcohol to disinfect the site upside down. Wait for 30 sec (allow the skin to dry)
4. Use Chlorhexidine / Povidone iodine to disinfect site concentric inside out. Wait for 1 min (allow the skin to dry)
5. Alcohol wipe to clean the blood culture bottle top
6. Collect blood aseptically & inject into blood culture bottle without changing the needle

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Conventional Culture Medium

Types of Media

1. Monophasic Medium
   - 50-100ml of Brain Heart infusion (BHI) broth
2. Castaneda's Biphasic Medium
   - BHI agar slope & BHI broth

Monophasic Medium

Castaneda's Biphasic Medium

- Dilution
  - Blood is inoculated in the medium at a dilution of 1:5 (1:10) so that the antibacterial components in the blood, if any, will get diluted

- Sodium polyanethol sulfonate (SPS)
  - SPS is added to the medium as an anticoagulant.
  - It also counteracts the bactericidal action of blood

- Incubation
  - Bottles should be directly incubated in upright position at 37°C for 7 days
  - Initial report comes in 3 days

- Repeat subcultures are made from the BHI broth onto Blood agar & Mac Conkey agar

Automated Culture Media

- BACTEC & BacT / ALERT
- BacT / ALERT Virtuo→Most advanced system
- Growth is continuously monitored and reading is recorded every 15-20 mins
- When growth is detected, the system gives a positive signal
- Then the bottle is removed and processed similarly as done for Conventional bottles

BACTEC

BacT / ALERT

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PrepLadder

Identification

- The isolated organisms is identified by
  - Colony morphology
  - Gram stain
- This is f/b either Conventional biochemical reactions or Automated identification system such as MALDI-TOF or VITEK

MALDI-TOF

VITEK

Antimicrobial Susceptibility Test (AST)

- AST is carried out for the guiding institution of appropriate therapy
- MIC based method (Eg. VITEK) is preferred over disc diffusion
- It is ideal for Endocarditis isolates, especially while reporting susceptibility of Penicillin

Treatment

- d/t high prevalence of Multi Drug Resistant Organisms (MDRO) & higher mortality in Sepsis, Antibiotics are started at earliest as soon as Sepsis is clinically suspected

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PrepLadder

Chapter 32

FEVER OF UNKNOWN ORIGIN

FUO
00:00:13

Despite doing intensive investigations not able to find out the real cause

Criteria
00:00:46

- Fever: 38.3°C on atleast 2 occasions
- Duration of illness: 3 wks
- No known immunocompromised state
- Diagnosis: uncertain diagnosis after History taking, Physical examination & various investigations

Investigation

- ESR + CRP
- Blood count (TLC, DLC, Hb, Platelets)
- Electrolytes, total proteins
- Enzymes
- ANCA & RF test
- Urine analysis
- Blood Culture (3 –ve Blood Cultures)
- Chest X ray, CT scan & MRI

Etiology
00:07:20

- Infections (36%)
- Neoplasms (19%)
- Non inflammatory diseases → SLE, RA
- Granulomatous disease
- Undiagnosed cases (7%)

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Systemic Bacterial Infections a/w FUO

00:09:15

- Mycobacterial infection
- Typhoid fever
- Rickettsial infection
- Melioidosis (B. pseudomallei)
- Listeriosis
- Bartonella
- Actinomycosis & Nocardiosis
- Syphilis
- Spirochete infection
  - Lyme's disease
  - Relapsing fever
- Leptospirosis

Viral Causes a/w FUO

00:13:52

- CMV & EBV
- Coxsackie virus (Group B infection)
- Viral hepatitis
- HIV infection
- Dengue
- Colorado tick fever

Parasitic Infections a/w FUO

00:15:25

- Malaria
- Amoebiasis
- Leishmaniasis
- Chagas disease
- Toxoplasmosis
- Echinococcus infection
- Schistosoma
- Toxocara canis/ catis

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Fungal infections a/w FUO

00:16:49

- Dimorphic fungus
- Aspergillosis
- Mucormycosis
- Candida & Crytpococcus
- Gram Staining → Pneumocystis pneumonia
  → Pneumocystis jiroveci pneumonia

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PrepLadder

Chapter 33

Anemia causing organisms

Iron deficiency Anemia

00:00:23

- T — Trichuris trichiura
- S — Schistosoma species
- H — Hookworm
  - Ancylostoma
  - Necator Americanus

Hemolytic Anemia

00:01:49

- B — Babesia
- B — Bartonella bacilliformis
- C — Clostridium perferingens
- England — EBV infection (Infectious mononucleosis)
- Main — Malaria
- Hai — Hep A

Megaloblastic Anemia

00:03:47

- Diphyllobothrium latum

Aplastic Anemia

00:04:18

- A
- P — Parvo B_{19}
- L — L. donovani
- A
- S
- T — TB
- I — Infectious mononucleosis (EBV)
- C — Hep C
- ANEMIA
- Hai — HHV_3, HHV_4, HHV_5, HHV_6 & HIV

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PrepLad

Chapter 34

Eye & Ear infections

Eye infections

00:00:13

External structures of Eye involved	Causative organisms
Eyelid (Blepharitis)	Staph aureus
Conjuctiva (conjuctivitis)	H. influenzae Chlamydia trachomatis Gonococcus Moraxella (Angular conjuctivitis) Adenovirus HSV
Cornea (Keratitis)	S. aureus Pneumococcus Fusarium Candida Acanthmoeba
Sclera (Scleritis)	S. aureus

Internal structures of eye involved

- Retina (Retinitis)
- Uvea (Uveitis)
- Aqueous humor or Vitreous Humor (Endopthalmitis)

Causative organisms

- Chorioretinitis & Uveitis
  - M.tb
  - T. pallidum
  - Borrelia burgdorferi
  - CMV
  - Toxoplasma gondii
- Endothalmitis
  - S. aureus
  - Pnemococcus
  - Pseudomonas aeruginosa

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- Other Gram Negative Bacteria (GNB)
- HSV
- Candida

Ear infections

00:09:03

Otitis externa / Swimmer's ear

- Acute Otitis externa
  - Causative organisms
    1. S. aureus (MC)
    2. S. pyogenes
    3. Pseudomonas
       - Common in DM
       - Causes Malignant Otitis externa
    4. Aspergillus
    5. Candida

Chronic Otitis externa

- Causative organisms
  1. Anaerobes (MC)
  2. Pseudomonas

Otitis media

Acute Otitis media	Chronic Otitis media
Causative organisms 1. Pneumococcus (33%) (MC) 2. Moraxella catarrhalis 3. S.pyogenes 4. RSV	Causative organisms 1. Anaerobes (MC)

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Chapter 35

Skin lesions

Primary skin lesions

00:00:20

1. Macule
   - Flat, non palpable discoloration of skin (<5mm in size)
   - Size >5mm → Patch
   - Caused by
     - Dermatophytes
     - T. pallidum (secondary syphilis)
     - Viral rashes (Enterovirus)

2. Papule
   - Elevated, palpable, sold lesion
   - Usually <5mm in size
   - Seen in
     - Molluscum contagiosum
     - Scabies
     - Warts (HPV)

3. Nodule
   - Elevated, palpable, solid lesion
   - Usually >5mm in size
   - Caused by
     - C.diptheriae
     - Sporothrix schenkii
     - PKDL
     - M. marinum
     - Nocardia
   - Size >2cm → Plaque

4. Vesicles
   - Fluid filled blister ≤5mm size
   - Caused by
     - HSV
     - VZV

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- M.contagiosum
- Hand, Foot & Mouth disease (Coxsackie A_{16})

5. Bulla
   - Fluid filled blister >5mm size
   - Caused by
     - C. perferingens (Gas Gangrene)
     - S. aureus (Bullous Impetigo)
     - Vibrio vulnificus (Halophillic bacteria)

6. Pustule
   - Pus filled lesion ≤5mm size
   - Caused by
     - Candida
     - S. aureus (Folliculitis)
     - S. pyogenes (Impetigo)
     - Dermatophytes

7. Abscess
   - Pus filled lesion >5mm size
   - Caused by
     - S. aureus
     - S. pyogenes

8. Petechiae
   - Blood extravasated into skin
   - Size <2mm
   - Caused by
     - HIV
     - Hep C
     - EBV
     - Rubella
     - CMV
     - Rocky mountain spotty fever (RMSF)

9. Purpura
   - Blood extravasated into skin
   - Size →2mm to 1cm
   - Seen in

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- CMV
- Endocarditis
- Meningococcemia
- Mononucleosis like syndrome
- Infectious mononucleosis
- RMSF
- Scarlet fever
- Sepsis
- Viral hemorrhagic fever

10. Ecchymosis
    - Blood extravasated into skin
    - Size >1cm
    - Caused by
        - Ebola virus
        - Marburg virus

      } Viral hemorrhagic fever

Other important skin infections

1. Scale
    - Excess dead epidermal layer
    - Caused by Dermatophytes

2. Ulcer
    - Break in epithelial lining extending upto epidermis / dermis
    - Caused by
        - Bacillus anthracis
        - M. leprae (Leprosy → Decubitus ulcer)

3. Erysipelas
    - Painful, red, indurated swollen lesion involving the dermis
    - a/w fever & lymphadenopathy
    - Caused by → S. pyogenes

4. Cellulitis
    - Diffuse spreading infection involving deeper layers of dermis
    - Caused by
        - S. pyogenes
        - S. aureus

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PrepLadder

5. Hidradenitis suppurtiva
   - Chronic infection of obstructed sweat glands
   - Caused by
     - S. aureus
     - Streptococcus anginosus

Hair follicle infections 00:19:59

1. Folliculitis
   - Superficial infection of single hair follicle
   - Mostly presents like Pustule
   - Caused by → S. aureus

2. Furuncle
   - Deeper infection of hair follicles
   - Presents as Abscess
   - Spreads deeply into the dermis & subcutaneous tissue
   - Caused by → S. aureus

3. Carbuncle
   - Coalescence of various furuncles
   - Caused by → S. aureus

Infection of Fascia & Muscles 00:24:06

1. Necrotizing fascitis
   - Rapidly spreading infection
   - Caused by → S. pyogenes (Flesh eating bacteria)

2. Pyomyositis
   - Pus formation in muscle layer
   - Caused by
     - S. aureus
     - S. pyogenes

3. Myonecrosis
   - Extensive necrosis of muscle layer with gangrene formation (Clostridial myonecrosis)
   - Caused by
     - C. perferingens
     - C. novyi

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PrepLadder

- C. septicum
- Other anaerobes

Agent causing Burn wound infections 00:27:31

1. S. aureus
2. P. aeruginosa
3. CONS (S. epidermidis)
4. Candida albicans

Agents causing Surgical site infections

1. Clean wounds
   - S. aureus
   - CONS
   - Enterococcus
   - Candida albicans

2. If Bowel integrity is compromised
   - Gram negative bacteria → E.coli
   - Anaerobic infections → Bacterioides, Prevotella

Lab diagnosis (in cases of skin & soft tissue infections) 00:32:21

Appropriate sample is take

- Pus taken up from wound → By Sterile swabs
- Pus taken up from abscess → By Incision & drainage
- Vesicles & Bulla fluid → By Needle aspiration
- Subcutaneous infections → Sample from base of lesion
- Dermatophyte infections
  - Skin scrapings
  - Nail clippings
  - Plucked hairs

Microscopy

1. Gram staining → Gives initial idea about Morphology of organism
2. 10% KOH mount for most of fungal infections
3. Tzank smear of vesicle fluid for HSV_1, HSV_2, HHV_3

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Culture

1. Aerobic organisms
   - Blood Agar
   - Mac Conkey agar

   } Incubate overnight at 37°c
2. Atypical Mycobacteria → LJ medium
3. Dermatophytes
   - DIM
   - SDA
4. Anaerobic organisms → Bacteria incubated anaerobically on
   - RCM (Robertson Cooked Meat Media)
   - BHI agar (Brain Heart Infusion Agar)

- Culture growth → go for AST (Antibiotic Sensitivity Testing)
  - System used for identifying organisms along with AST patterns → Vitek
  - Vitek also tells about MIC

Vitek

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PrepLadder

Chapter 36

Nosocomial Infections / Hospital Acquired Infections (HAI)

Definition → Infection occurring >2days of Hospital admission

Classification

00:00:34

- CAUTI → Catheter associated UTI
- CRBSI → Catheter related blood stream infection
- VAP → Ventilator associated Pneumonia
- SSI → Surgical Site Infections

CAUTI

00:02:17

- MC HAI worldwide
- Short term catheterization
  - Causative organisms
    - E.Coli (MC)
    - Klebsiella
    - Pseudomonas
    - Enterococci (GPC)
- Long term catheterization
  - Usually polymicrobial infection occurs
  - Causative organisms
    - Pathogens of short term catheterization + PMP

      P → Proteus

      M → Morganella

      P → Providencia

Source of infection

Extra luminal spread	Intra luminal spread
Mostly endogenous flora or Soiled hands of Healthcare workers	Open drainage bag (allows organisms to enter & cause infection)

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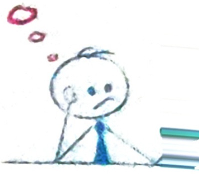

Lab diagnosis

Catheter criteria	Catheterized or history of recent Catheterization within 48hrs
Clinical criteria	Presence of at least one sign or symptom of UTI - Fever - Suprapubic tenderness - Costovertebral angle pain - Urinary urgency, frequency or dysuria
Urine culture criteria	Presence of significant bacteriuria; defined as Colony count exceeding: - $\geq 10^3$ CFU/mL: in symptomatic patients - $\geq 10^5$ CFU/mL: in asymptomatic patients

CRBSI

00:07:54

- a/w Central venous catheter or Central line
  - Temporary or short term central line (<72 hrs)
  - Permanent or long term central line (>72 hrs)
- Site of insertion of Central line
  - Subclavian vein
  - Femoral vein
  - Internal Jugular vein
  - Peripheral veins
- Organisms gain entry
  - d/t colonization of organisms on Central line
    eg. CONS (Biofilm), S. aureus

Clinical criteria for CRBSI

- Presence of fever, chills, rigor, hypotension after central line is used

or

- Signs of catheter site infection present
  - Erythema, Tenderness, Swelling at the catheter exit site

Microbiological criteria for CRBSI

- Take blood culture from both central line as well as peripheral line
- If central line blood culture bottle flags ≥2 hrs prior to peripheral line blood culture bottle

↓

Indicative of CRBSI

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VAP

00:14:25

- 2nd MC nosocomial infection after CAUTI
- Accounts for 15-20% of total HAI's
- MCC of death among HAI (Mortality rate →40%)

Early onset VAP

- Occurs in first 4 days of Mechanical ventilation
- Caused by
  - Pneumococcus
  - H. Influenzae
  - MSSA (Methicillin Sensitive Staph aureus)

Late onset VAP

- Develops ≥5 days after Mechanical ventilation starts
- Caused by
  - P. aeruginosa
  - Acinetobacter baumanii
  - E. coli
  - Klebsiella
  - MRSA (Methicillin Resistant Staph aureus)

Source of infection

- Endogenous flora

  or
- Exogenous infection
  - from Heathcare workers, Hospital environment

Clinical criteria for VAP

- CPIS score >6 →Diagnostic of VAP

  * CPIS →Clinical Pulmonary Infection Score

Microbiological criteria for VAP

- Specimens taken
  - Endotracheal aspirate (ETA) →MC
  - BAL (Broncho Alveolar Lavage) fluid
  - Lung biopsy (by Bronchoscopy)

- Do Gram staining & Culture
  - Quantitative assessment on Culture

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- ≥10^5 CFU/mL for ETA
- ≥10^4 CFU/mL for BAL

- Semiquantitative assessment on Culture
  - Moderate to Heavy growth

Radiological investigations

- CT Scan
- CXR

SSI

00:21:30

- Infections which develop within 30 days of surgery at surgical site

- Exception
  - Breast Sx
  - Cardiac Sx
  - Joint Sx

  } ≥90 days of Surgery

Source of infection

- Endogenous flora

  or

- Exogenous infection
  - from Heathcare workers, Hospital environment

Wound class types

- Class I wounds → Clean wounds
- Class II wounds →Clean contaminated wounds
- Class III wounds
  - Contaminated wounds →Mostly a/w accidents
  - Dirty & infected wounds → Surgical procedure done over active infection

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Other classification of SSI

Superficial SSI	Deep SSI
Occurs at level of superficial incisional site (Skin & Subcutaneous tissues)	Occurs at level of deep incisional site (Muscle & Fascia)
Happens within 30 days of surgery	Happens within 30 days of surgery Exception - Breast Sx - Cardiac Sx - Joint implant Sx } ≥90 days of Surgery

Prevention of SSI

- Pre operative measures
  - Pre operative bathing
  - Treat MRSA carriers with Mupirocin ointment
  - Hair removal should not be done. Hair clipping is done now a days

- Intra operative measures
  - SAP (Systemic Antibiotic Prophylaxis)
  - Surgical hand disinfection by best possible ways
  - Proper surgical site preparation
  - Proper Oxygenation (Target $FiO_2 \rightarrow 80\%$)
  - Maintain normal blood glucose levels during Sx (Target <200mg/dL)

- Post operative measures
  - Proper wound dressing → Done daily
  - OT disinfection
  - Periodic monitoring of air quality in OT

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PrepLadder

Chapter 37

Hand hygiene

- Best method to prevent Nosocomial infections

Hand Rub 00:00:41

- Done with
  - Alcohol based Rubs (70-80% Ethyl alcohol)
  - Chlorhexidine
- Duration → 20-30 seconds

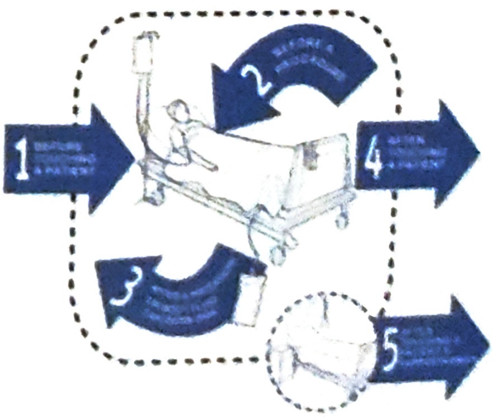
Hand Wash 00:01:30

- Done with Antimicrobial soaps
- Duration → 40-60 seconds
- When to do hand wash
  - When hands are visibly soiled with blood, pus or excreta
  - Before & after eating
  - After going to toilet
  - Before and after duty (in case of Healthcare personnel)

Surgical hands scrub 00:03:37

- Done with 4% Chlorhexidine
- Duration → 3-5mins
- Done prior to any surgical procedure

Five moments for Hand Hygiene (as per WHO) 00:04:25



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Steps of Hand Rubbing & Hand Washing   00:05:37

Step 1 → Apply Hand Rub / Hand Wash
Step 2 → Palm to Palm
Step 3 → Back of the palm
Step 4 → Interlacing of fingers
Step 5 → Back of the fingers
Step 6 → Rotational rubbing of thumb
Step 7 → Scrub nails on palm
Step 8 → Rinse the hands with water
Step 9 → Dry hands with a single use paper towel
Step 10 → Close the tap with (Elbow > same single use paper towel)

Personal Protective Equipments (PPE)   00:09:04

1. Gloves
2. Heavy duty gloves
3. Surgical mask
4. N_{95} respirator
5. Plastic apron
6. Linen gown
7. Disposable gown
8. Coverall
9. Goggles
10. Face shield
11. Cap
12. Shoes

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PrepLadder

13. Gum boot
14. Shoe cover

Donning → Wearing PPE 00:11:50

- Steps of Donning
  1. Gown
  2. Mask / N_{95} respirator
  3. Goggles / face shield
  4. Gloves

Doffing → Removing PPE

- Steps of Doffing
  1. Gloves
  2. Goggles / Face shield
  3. Gown
  4. Mask / N_{95} respirator

Blood Spill Management 00:16:08

- Blood spill management is needed to remove the risk of transmission of blood borne viruses
- Steps to follow if a blood spill happens
  - Contain the spill and put a Signage at the place where spill happened
  - Call Infection Control Team
  - Sodium hypochlorite used in management of blood spill should be freshly prepared

As per CDC guidelines

a. For large spills → ≥10cm size
   - Use Sodium hypochlorite → 0.5% (1%)

b. For small spills → <10cm size
   - Use Sodium hypochlorite → 0.05%

c. Contact time → 10 mins

d. Rinse the area with clean water to remove the disinfectant residues

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Chapter 38

Organisms transmitted by Droplets and Aerosol

Respiratory droplet 00:00:13

- Large particles >5μ in size
- Generated by a person while coughing, sneezing or talking
- Transmission doesn't happens beyond 3 feets

Infections spread by droplets 00:01:32

MDH → Meningococci, Diptheria, Hib

in

PPM → Pertussis, Pneumonic Plague, Mycoplasma pneumonia

IS → Influenza virus, SARS COV2 (COVID 19)

Very → Viral Hemorrhagic fever (Ebola, Marburg, Lassa virus)

Owesome → Other viruses

- A- Adeno virus
- R- Rhino virus, Rubella
- M-Mumps
- P- Parvo virus B_{19}

Aerosols → Particles <5μ in size 00:06:00

Infections spread by Aerosols

M - M.tb

M - Measles

S - Small pox

A - Aspergillus

B - Bacillus anthracis

C - Chicken pox

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Chapter 39

Biosafety levels

- Level of biocontainment precautions required to isolate dangerous biological agents in enclosed facility

BSL – 1 00:01:26

- Biological agents pose low risk to personnel & environment

  Big →Bacillus thuringiensis
  P →Pseudomonas fluorescens
  E →E.coli K_{12}
  A →Aspergillus niger
  S →Serratia marcescens

- BSL-1 is found in
  - Water testing labs
  - High Schools & College teaching introductory Microbiology classes
- Work is done on an open bench
- Lab coats & gloves are recommended

BSL-2 00:04:20

- Biological agents pose moderate risk to personnel & environment
- Risk of spread is limited & it rarely would cause infection that would lead to serious disease

- Eg. For BSL-2
  - Bacteria→ Pathogenic Staph, Streptococci, Pneumococci, Clostridium species, Bacillus (except Anthrax), Enterobacteriacea, Pathogenic E.coli

  - Viruses → Herpes, Calci, Influenza, Adeno (HCC Is Aggressive)
  - Fungi → Aspergillus, Dermatophytes, Candida, Cryptococcus (ADCC)
- Diagnostic work of COVID 19
  - PCR / Tru NAT → Done in BSL-2

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BSL-3

00:07:32

- Biological agents usually produce serious disease
- Lab must be in a separate building or isolated zone with double door entry, directional inward flow.
- Culture of SARS COV 2 (COVID 19) (for vaccine production) is done in BSL-3
- Eg. of BSL-3
  - Bacteria
    - A → Anthrax
    - B → Brucella
    - C → Coxiella burnetti
    - T → Tularemia
    - Y → Yersinia pestis
    - M → M.tb
  - Viruses (Arboviruses) → Hantan virus, Lymphocytic choriomeningitis virus, St. Louis encephalitis virus, Japanese encephalitis virus, West nile encephalitis virus, Chikungunya, Yellow fever virus
  - Fungi (Dimorphic fungi) → Coccidiodes immitis, Blastomyces dermatidis, Paracoccidiodes brasilensis, Histoplasma capsulatum

BSL-4

00:12:51

- Biological agents pose very serious infection that is often untreatable
- Easily transmitted from one to another individual, from animal to human and vice versa either by direct, indirect or by casual contact
- Eg.
  - Lassa fever virus
  - Marburg virus
  - Ebola virus
  - CCHF (Crimean Congo Hemorrhagic Fever) virus
  - Hendra virus
  - Nipah virus

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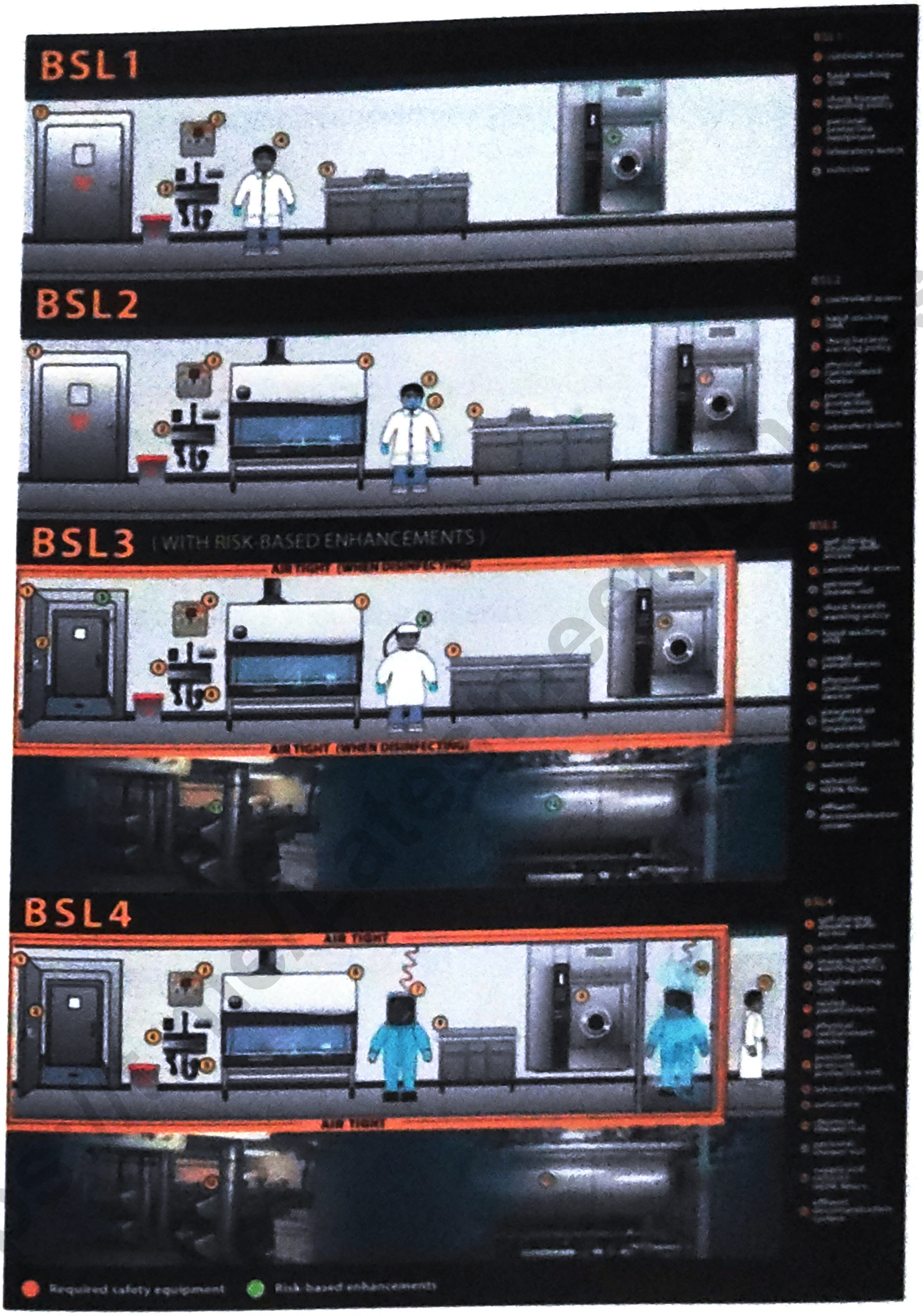
BSL1
BSL2
BSL3 (WITH RISK-BASED ENHANCEMENTS)
AIR TIGHT (WHEN DISINFECTING)
AIR TIGHT (WHEN DISINFECTING)
BSL4
AIR TIGHT
AIR TIGHT
Required safety equipment
Risk-based enhancements

Chapter 40

Occupational Exposures (HIV & HBV)

00:00:20

1. Percutaneous injuries
   - Needle stick injury / Sharp injury
2. Splash injury
   - Contact with mucus membrane
   - Contact with non intact skin
   - Contact with intact skin

Needle Stick Injury

00:02:05

Agents	Rate of transmission
HBV	30%
HCV	3%
HIV	0.3%

Potential infectious body fluids

00:03:15

- Blood
- Genital secretions
- All body fluids → CSF, Synovial fluid, Pleural & Peritoneal fluid

Not considered potentially infectious

- Faecal matter
- Nasal secretions
- Saliva
- Sputum
- Sweat
- Tears
- Urine
- Vomitus

Precautions during needle handling

00:06:41

1. Work surface → Disinfect with 0.5% Na hypochlorite
2. Healthcare workers – Immunized against HBV

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3. If spillage of blood / body fluids happen
   - Mx with 1% Na hypochlorite
4. Disposable needle should be used (No Recapping)

Post exposure management
00:09:50

1. First aid
2. Report to Nodal center
3. First dose of Post Exposure Prophylaxis (PEP) within 2hrs (HIV) & at the most before 72hrs
4. Test for other blood borne viruses
5. Record the incident
6. Follow up testing of Healthcare worker
7. All precautions should be taken during follow up

First Aid
00:13:18

- For Splash injury
  - Irrigate thoroughly the site vigorously for atleast 5mins
  - In oral contact → spit fluid immediately & rinse the mouth several times
  - If wearing contact lens
    - Leave them in place
    - Eye is cleaned first
    - Remove contact lens in normal fashion

Don'ts
00:16:08

1. Don't panic
2. Never put pricked finger in mouth
3. Don't squeeze finger

PEP for HIV as per NACO guidelines
00:17:25

Exposure Codes (EC)

EC-1 (Mild Exposure)
- Mucous membrane contact / non intact skin contact with small volume of blood / fluid or less duration

EC-2 (Moderate Exposure)
- Mucous membrane contact / non intact skin contact with large

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volume of blood / fluid or more duration

- Percutaneous superficial exposure with solid needle or just a superficial scratch

EC-3(Severe Exposure)

- Percutaneous exposure with
  1. Large volume transfer
  2. Deep puncture of needle
  3. Visible blood on device
  4. Needles used in blood withdrawing from arteries & veins

Source Codes (SC)

SC-1

- HIV positive but asymptomatic
- Viral load <400 copies /ml

SC-2

- HIV positive, asymptomatic
- High viral load

SC unknown

- Status of patient is unknown

HIV negative

- Tested negative according to NACO strategies

Exposure Code	Source Code	PEP
1,2 or 3	Negative	Not needed
1	1	Not needed
1	2	Needed
2	1	Needed
2	2	Needed
3	1 or 2	Needed
2/3	Unknown	Needed

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PEP

- Started within 2hrs
- It is of no use if started after 72hrs
- Duration → 28 days

PEP Regimen

- Tenofovir
- Lamivudine

} 1 tablet daily x 28 days

- Lopinavir
- Ritonavir

} 2 tablets twice daily x 28 days

Alternative Regimen → TLE

- Tenofovir → 300mg
- Lamivudine → 300mg
- Efavirenz → 600mg

1 tablet daily x 28 days

PEP for Hep B

00:30:55

Healthcare worker status	Source is positive (HBs Ag +ve)	Source is negative (HBs Ag -ve)
If exposed person is completely vaccinated & Ab titer >10 mIU / ml	No further treatment required	
If Ab titer of exposed person <10 mIU / ml (vaccinated person)	Give HBIg dose immediately (max. upto 7 days) Start 2nd series of vaccine	Start 2nd series of vaccine
If exposed person is partially vaccinated or not vaccinated	Give HBIg dose Complete remaining vaccination	Complete remaining vaccination
Vaccinated for 2 series still Ab titers <10 mIU/ml (Non responders)	Give HBIg 2 doses 1 month apart (0.06ml / kg) or (10-12 IU/kg)	Nothing required

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Chapter 41

Biomedical Waste Management

- Any waste generated during lab diagnosis, treatment or immunization, research activities

Waste generated in hospitals

00:01:04

1. General waste = 80%
2. BMW = 20%
   - 10% Infectious waste
   - 5% Chemical / Radioactive waste

Aim for waste management

00:02:24

- Reduce
- Recycle 3'R'
- Recover

BMW guidelines 1998

- New guidelines given by Ministry of Environment & Forests
- Given in 2016 & amended in 2018-19

Steps of BMW management

00:04:17

1. Waste segregation → At point of generation
2. Pretreat → Lab liquid waste
3. Transportation of waste
   - From generation site → Central storage area (CSA)
   - From CSA → Common Biomedical Waste Treatment Facility (CBMWTF)
   - Waste should reach from generation site to Common Biomedical Waste Treatment Facility within 48hrs
4. Treatment and disposal of waste in 48 hrs

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PrepLadder

00:07:27

BMW guidelines

Category	Type of waste	Type of Bag / Container	Treatment / Disposal options
Yellow (Infectious non-plastic waste)	Human anatomical waste	Yellow coloured non chlorinated plastic bags	Incineration / Plasma pyrolysis / Deep burial
	Animal anatomical waste		
	Soiled waste		Incineration / Plasma pyrolysis / Deep burial / Autoclaving or Hydroclaving + Shredding / Mutilation
	Expired / discarded medicines-Pharmaceutical waste, Cytotoxic drugs	Yellow colored containers / non chlorinated plastic bags with Cytotoxic label	Sent back to manufacturer/ CBMWTF for Incineration (Cytotoxic drugs at temperature ›1200°c)
	Chemical solid waste	Yellow colored containers / non chlorinated plastic bags	Incineration or Plasma pyrolysis or Encapsulation
	Chemical liquid waste such as discarded disinfectants, infected body fluids and secretions, liquid from house - keeping related activities	To be discharged into separate collection system, which leads to effluent treatment system Not to be discarded into yellow bag	Pre -treated before mixing with other waste water
	Discarded linen waste contaminated with blood / body fluids, mask, cap, gown and shoe cover	Non - chlorinated yellow plastic bags / suitable packing material	Non - chlorinated chemical Disinfection followed by Incineration / Plasma pyrolysis
	Microbiology, other clinical laboratory waste, blood bags, live / attenuated vaccines	Autoclave safe plastic bag / container	Pre-treat to sterilize with non -chlorinated chemicals / on-site as per NACO / WHO guidelines + Incineration

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Red (Infectious plastic waste)	Disposable items such as tubing,bottles, intravenous tubes and sets, catheters, urine bags, syringes (without needles and fixed needle syringes) and vacutainer with their needles cut, gloves, plastic apron	Red colored non-chlorinated plastic bags or containers	1. Autoclaving / Microwaving / Hydrovlacing+ Shredding 2. Mutilation / Sterilization + Shredding 3. Treated waste sent to authorized recyclers or for energy recovery
White (Translucent) (Waste sharps including metal sharps)	Needles, syringes with fixed needles, needles from needle tip cutter or burner, scalpels, blades, or any other contaminated sharp (used or discarded)	Puncture-proof, leak-proof, tamper-proof containers	Autoclaving / dry heat sterilization followed by: 1. Shredding or Mutilation or Encapsulation in metal container or cement concrete or 2. Sanitary landfill or 3. Designated concrete waste sharp pit
Blue (Glasswares & metallic body implant	Broken, discarded & contaminated glass including medicine vials & ampoules except those contaminated with cytotoxic waste	Blue colored containers	

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BMW Treatment Modalities 00:21:02

1. Incineration
   - Dry heat sterilization method
   - High temperature →800-1200°C
   - Organic waste is converted into non organic waste
   - Waste volume is decreased
2. Autoclave
   - Moist heat sterilization method
   - Used for Infectious plastic waste & sharp waste
3. Chemical disinfection
   - 1-2% Sodium hypochlorite
4. Microwaving
   - Radio frequency waves of 2450 MHz are used

     ↓

   Produce friction of water molecules

     ↓

   Generation of heat
5. Hydroclaving
   - Low temperature steam method
   - Waste is broken into small pieces
   - Done to avoid Shredding
6. Shredding
   - Waste is cut into small pieces
   - Helps in decreasing waste volume
7. Deep Burial
   - Pit is dug at 2 metres depth
   - Pit is half filled with waste & then covered with lime within 50cms of surface before filling rest volume with soil

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8. Sharp Pits
   - Constructed within Hospital
9. Encapsulation
   - Immobilizing the material & sealing up the containers
10. Inertization
   - Mixing waste with cement before disposal
11. Plasma Pyrolysis
   - Ionized gas is used in Plasma state
   - Costly method
12. Expired Cytotoxic waste
   - Resent back to manufacturer

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OWN YOUR DREAM

I am Dr. Shivani H. Patel from Gujarat, India. My story is not an extraordinary one but somehow, I felt the need to share it. I feel it can inspire some of my peers to never give up, to believe in their dreams and carry on.

2020 NEET PG was approaching; my preparation was in full swing. I was aiming to achieve my dream and was preparing with zeal and enthusiasm to bag a seat in General Surgery. In November, I started experiencing pain in my right leg; it was so bad that I had to consult a doctor. He prescribed me some pain killers, but even after 15 days of regularly taking the medicines, the pain persisted. I was not able to move or even sit still for over 10 minutes. This was the peak time for preparation and while everyone was spending hours studying at a stretch, I was struggling to even sit.

After another visit to the doctor, I was diagnosed with Sciatica. The prescribed drug 'Gabapentin' had me feeling drowsy all day, due to which my studies were suffering. So, I decided to stop the medication, which I know was wrong, but I wanted to focus on my studies. I would keep pillows on the sides and study while lying down. That's how I gave my NEET PG 2020 exam. The results came and I didn't score enough to get my dream branch. I was disappointed but I realised health is above everything.

I got my MRI done and was diagnosed with L4 – L5 disc bulge, compressing nerves accompanied by severe pain. I am doing physiotherapy and taking regular medication this time. I recently suffered a neck sprain and nerve compression too but I am not ready to give in.

My friends and colleagues told me to go for a Non-Clinical branch, but I am adamant on what I want, so I've decided to give NEET PG another shot. I am now preparing for 2021 NEET PG and I hope I can make it.

I want to give my best to achieve my dreams, because giving up is easy but going on takes guts. I believe the ones who persist are the ones who win. So, let's keep persisting. Let's never give up on our dreams.

Dr. Shivani H. Patel

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Trust in dreams, for in them is hidden the gate to eternity.

- Khalil Gibran

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